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# **Quarantine Security for Commodities: Current Approaches and Potential Strategies**

**Proceedings of Joint Workshops  
of the Agricultural Research  
Service and the Animal and  
Plant Health Inspection Service**

**June 5—9 and  
July 31—August 4, 1995**





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**Nicanor J. Liquido, Robert L. Griffin,  
and Kenneth W. Vick, editors**

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## ABSTRACT

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This publication discusses the relationship of the Animal Plant Health and Inspection Service (APHIS) and the Agricultural Research Service (ARS) to the management of pest and disease threats to plant resources. APHIS and ARS regulators and scientists evaluated existing and potential alternatives for characterizing quarantine security, which includes probit 9. Results are summarized in this series of papers. The objective is to prove that the concept of probit 9 is particularly effective for treating high-risk insect pests, such as fruit flies, although probit 9 is not used for all quarantine security conditions.

**Keywords:** pest-free areas, pest risk assessment, probit 9, quarantine security.

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## Foreword

The Animal and Plant Health Inspection Service (APHIS) and the Agricultural Research Service (ARS) have a long history of cooperation in matters related to the management of pest and disease threats to plant resources. This document reflects the latest evolution of that relationship by discussing contemporary positions developed by APHIS and ARS on the key issue of quarantine security.

The term “quarantine security” has a degree of ambiguity associated with it. Because no precise definition exists for the term, the concepts of risk, acceptable risk, and affordable risk continue to be debated in national and international forums. The discussions herein are neither designed to intensify the debate nor precisely define any of the above concepts. Rather, this represents an effort by the USDA to identify and describe methods for achieving the goals of pest risk management in various ways.

Historically for USDA, quarantine security has been synonymous with “probit 9,” or 99.9968 percent mortality. Probit 9, in spite of its critics and recognized limitations, has proven to be an effective tool for many situations, particularly the approval of treatments designed for high risk insect pests such as some fruit flies. However, the extensive use and citation of this concept has led to the assumption that probit 9 is the USDA’s adopted benchmark for quarantine security under all conditions. Clarification of this point has been identified by APHIS and ARS as an important objective.

Through a series of meetings and ongoing consultation, regulators and scientists from APHIS and ARS evaluated a range of existing and potential alternatives for characterizing quarantine security, including the role of probit 9. The series of papers assembled here represent the results of these efforts summarized in terms of separate concepts.

The document begins by placing the existing concepts into perspective. Probit 9 is discussed here alongside treatises on Pest Free Areas and Systems Approaches. This is followed by a section devoted to potential strategies. Alternative Levels of Mortality and the Maximum Pest Limit concepts are described in this section.

Each concept begins with an introduction and definition to bring the concept into focus. Applications are then presented with selected examples, and data or other requirements are then discussed. Opportunities and limitations follow together in order to allow direct comparison of the advantages and disadvantages of the concept. Finally, a brief treatment of the quantitative and qualitative elements contributing to the application of the concept is presented.

Biometrics associated with risk assessment of each concept follows the section on opportunities and limitations. In addition, software has been developed by ARS and is included in an appendix to facilitate the comparison of important calculations: 1) a SAS program used for all biometrical concepts and formulae presented; 2) a user-friendly application designed to allow scientists and regulatory officers without adequate statistical expertise to run risk estimates as defined in Section II: Potential Strategies.

All appropriate references are summarized in a separate section.

In sum, the material presented here serves the dual role of clarifying current positions while identifying future options. It is hoped that this will assist those wishing to understand the complexity of quarantine security issues and also stimulate a continuing dialogue on quarantine security in the regulatory and scientific community at large.

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**Proceedings of Joint Workshops of  
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## SECTION I. Current Approaches

### Probit 9 Efficacy

#### Introduction

Commodity treatments for pests requiring a high degree of quarantine security are commonly referred to as probit 9 treatments. The reference originates from the statistical method (probit analysis) used for deriving the dose-response relationship. This method may be used for predicting the level of treatment needed to achieve the desired degree of response or the efficacy of a given treatment (dose).

A response at the probit 9 level results in 99.9968% efficacy. The response may be mortality, sterility, or preventing the maturation of pest organisms.  $LD_{99.9968}$  is a less ambiguous term when discussing probit 9 in terms of mortality. However, lethal stimulus (LS), lethal time (LT), lethal concentration (LC), and a general mortality factor ( $Q$ ) have also been proposed as more appropriate terms to describe responses when the criterion is mortality (Robertson, et al. 1994).

The USDA has used 99.9968% mortality as the basis for approving many quarantine treatments since it was described by Baker in 1939. However, the concept has not endured without some scrutiny. Discussing the concept in 1984, Landolt states, "There appears to be no reported or published discussion or account of reasons for using mortality rates as criteria for quarantine disinfestation treatments, or for setting the required mortality rate at probit 9." (Landolt et al. 1984).

Landolt goes on to explain that quarantine security is not adequately measured by considering mortality alone. He makes the point that a high mortality rate serves to lower the pest risk, but not necessarily to within reasonable or acceptable limits. The implication is that the risk

of pest introduction is more than a function of mortality, but involves variables not considered when using probit 9 as the sole criteria for gauging quarantine security. Herein lies one of the primary criticisms of the concept.

While probit 9 is clearly designed to reduce the prevalence of pests by a predictable amount, it does not account for other variables contributing to the pest risk. Natural survival rates, the likelihood of infestation, and the colonization potential of the pest are a few of the more important risk-based considerations that are ignored by a direct estimation of mortality such as probit 9. Process parameters such as pre-shipment cultural practices, packaging and shipping procedures, and distribution times or areas are not considered when mortality is the sole criterion for determining quarantine security.

Nevertheless, probit 9 has enjoyed widespread use over many years, resulting in the impression that probit 9 is the universal standard for quarantine security regardless of the pest risk, and that the probit model is the only valid statistical device for demonstrating treatment efficacy. These assumptions are supported by the proportionally larger number of USDA approved quarantine treatments based on probit 9 efficacy. However, this ignores the range of alternative analytical techniques and mitigation options upon which many other USDA authorizations are based. Multiple or combination treatments, pest-free areas, systems approaches, and a variety of specially designed inspection schemes have also provided the basis for establishing quarantine security. In addition, many different analytical or statistical devices have been found acceptable by ARS and APHIS for evaluating data -- but none have the renown of probit 9.

Establishing the level of quarantine security at probit 9 for single treatment disinfestation options was described by Baker as having two objectives: 1) to assure essentially complete efficacy against target organisms found in treated commodities;

and 2) to lighten the treatment requirement as much as possible to reduce damage to the commodity.

Developing a treatment and demonstrating probit 9 efficacy frequently proves to be the quickest and most easily accepted method for overcoming phytosanitary restrictions. The rigorous nature of a probit 9 treatment virtually assures pest freedom for normal commercial shipments without the need to evaluate other more complex, tenuous, and potentially data-intense variables associated with the risk of pest introduction.

### Requirements

The mathematical derivation of a dose-mortality relationship is based on the assumption that each organism or experimental unit (e.u.) has a tolerance for some stimulus (dose, temperature, time, etc.). The e.u. will respond if it receives a stimulus exceeding its tolerance threshold. Probit is a mathematical transformation of the proportion for dose-response (e.g., the number of organisms that are killed out of a total population). If the tolerance of the organisms is distributed normally, the regression of probit on dose will be linear. If the distribution is lognormal, the regression of probit on log (dose) will be linear. Probites of proportions which are less than 0.5 are negative. Finney (1971) proposed adding five to the probit transform of the proportion to ensure that probit values are always positive.

Two closely related transformations that are in common use are the logit and the complementary log-log. The regression of the logit and the complementary log-log transforms on dose will be linear if the distribution of the tolerances is logistic or Weibull, respectively.

Baker assumed that mortality and the length of exposure were related in a linear fashion using a lognormal transformation. However, dose-response data are not always distributed normally, and there is no theoretical or experimental way to

determine the tolerance distribution. Therefore, a goodness-of-fit test should always precede the selection of the statistical device in order to determine if probit is appropriate. In many cases, two or more models may demonstrate no lack of fit, in which case the choice of a suitable model must be based on other (subjective) criteria.

The requirement that quarantine security meet probit 9 criteria is most frequently employed where the following circumstances exist:

- the host is suitable;
- the infestation rate is high;
- the host is easily infested;
- distribution of the pest is highly clumped; and
- the pest is internal or difficult to detect.

Other important factors relate to the susceptibility of the commodity and the pest to the proposed treatment. A probit 9 level treatment is more likely to be nominated in situations where the treatment has a strong effect on the pest and little effect on the quality of the host. In addition, demonstrating probit 9 level efficacy may be preferred over other methods when pest organisms can be easily collected, maintained, and treated in large numbers.

### Applications

Contemporary examples of currently approved treatments that were developed using the pure application of probit 9 include the following two which were developed to establish quarantine security for fruit flies in mangos:

#### Mango-Vapor Heat

This treatment is approved by APHIS for the treatment of mangos for *Anastrepha ludens* (Mexican Fruit Fly) (Mangan and Ingle 1994). The treatment authorization schedule is listed as T-103(a) in the PPQ Treatment Manual.

#### Mango-Hot Water

The hot water dip treatment for mangos is one of



the most successful non-chemical treatments developed for quarantine purposes. Data required to expand the geographical scope of the treatment from Mexico alone to nearly all Western hemisphere countries was based on the work of Sharp and Picho-Martinez (1990). The corresponding PPQ Treatment Manual listing is schedule T-102(a).

### **Opportunities and Limitations**

Criticism for the inadequacies of probit 9, especially the extreme and inconsistent level of quarantine security it provides, has led to its falling in disfavor among many treatment experts (Robertson et al. 1994). However, it remains an important concept for regulators in a less technical framework because it is quickly identified with a high pest risk situation requiring rigorous mitigation. Although it is unlikely that probit 9, in the strictest sense of the concept, will flourish as a means for establishing quarantine security, it has several advantages which are likely to keep it within the mix of tools used for establishing quarantine security.

Major advantages are:

- The concept is easily identified with high risk organisms.
- It provides an alternative to more arduous data collection related to variables associated with estimating the risk of pest introduction and establishment.
- A conservative, albeit inconsistent, level of quarantine security is achieved.
- It provides a means for comparison with other, new, or different schemes for establishing quarantine security.

The limitations associated with adopting probit 9 as the means for achieving quarantine security are many. Some important limitations are identified in the discussions herein. The following is a

summary listing some of the major points of contention:

- Probit 9 is an arbitrary level and probit analysis has not been proven to be statistically appropriate for the analysis of bioassay data.<sup>1</sup>
- Transformation is not always necessary for dose-response data to be linear.<sup>2</sup>
- 99.9968% efficacy, or any other very high level of efficacy, is difficult to estimate with a high degree of certainty because these values reside in the extreme upper end of the probability distribution.
- Probit analysis assumes that response data are distributed normally and that transformation is necessary to express dose-response data in a linear fashion.
- Fitting probit lines to mortality data for a fixed dose with time replacing dose as a variable can be difficult if not using different batches of organisms for different times.

### **Pest Free Areas**

A viable alternative for exporting countries to address quarantine requirements for the entry of fresh agricultural products in lieu of commodity treatment would be to establish and maintain specifically defined areas or regions free of certain pests for a given commodity or group of commodities.

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<sup>1</sup> Probit analysis is exact if the assumption of normal distribution of the tolerance is correct. Any other analysis (e.g., logistic or logit analysis) must also assume a distribution of tolerances.

<sup>2</sup> All transforms result in the prediction of mortality between 0 and 100%. Without transformations, it is possible to predict negative mortality at low doses and mortality exceeding 100% at high doses.

The pest free area (PFA) concept is a risk management option based upon a sound pest risk assessment coupled with satisfactory evidence of effective, on-going surveillance and exclusion measures to maintain such areas pest free.

PFA status has been successfully used by an increasing number of countries to issue phytosanitary certificates to permit the movement of regulated plants and plant products in international commerce that otherwise would have required treatment or may have been prohibited.

### **Definition of Pest Free Area**

For purposes of this paper, a pest free area is defined as "An officially identified area in which a target pest does not occur and is maintained as such."

PFA status does not imply an absolute absence of all pests, but is aimed at designated commodities from specific geographic areas based on the absence of a specific pest or group of pests.

PFAs are based on the absence of a target pest or complex of pests. It is not synonymous with areas of low prevalence of a target pest or pests. Areas of low prevalence more appropriately could be considered for phytosanitary certification under a systems approach relying upon a combination of pest management practices and other mitigation measures to reduce risk to an acceptable level.

### **Concepts Associated With Pest Free Areas**

Recognition of areas as "pest free" is not a totally new concept. The United States Department of Agriculture (USDA), Animal and Plant Health Inspection Service (APHIS) has determined entry status for many imported products based on the presence or absence of quarantine pests of concern in the country of origin. In most instances, these assessments are based on an extensive review of scientific literature, knowledge of the distribution of the pest in question, and results obtained from agricultural

quarantine inspection such as records of interceptions or the lack of interceptions. Conversely, other countries have permitted entry of U.S. products based on USDA certification of pest absence.

Although recognition of pest free status is based upon an initial pest risk assessment to determine what pest or pests may be present, PFAs are oriented toward risk mitigation and risk management. PFAs set forth phytosanitary procedures that allow plant or plant products to move in commerce rather than impose restrictions on such movement, except as it pertains to maintaining these areas pest free.

All pest free zones officially recognized to date by the USDA have involved exotic species of fruit flies. Existing regulations (as cited in Title 7 of the United States Code of Federal Regulations, Part 319 -- Foreign Quarantine Notices, Subpart - - Fruits and Vegetables, Section 319.56-2 Restrictions on entry of fruits and vegetables) apply only to areas free from injurious insects, including fruit and melon flies (Tephritidae). USDA now considers PFAs in a much broader context that potentially could be extended to other plant and animal pests and diseases, and considered on a regional basis that could include an uninfested area of a country, a whole country or portions of several countries.

By regulation, the PFA concept applies to situations where a specific area or district of a country has been found completely free of a pest or is made free through specific actions, and is then protected from infestation or re-infestation. In practice, PFAs have involved clearly defined geographic boundaries and the degree of geographic isolation from the nearest infested areas has contributed significantly to their successful establishment.

Although the regulations do not specify a minimum size, the practicality of applying PFA to greenhouses or premises is questionable in terms



of effectiveness of exclusion measures and sensitivity of surveillance/detection. Since the issue would rest on low prevalence rather than complete absence, these sites would be addressed more appropriately under a systems approach.

### **Current Policies**

Current USDA regulations are cited in 7CFR319.56-2 and apply to almost any fruit (except citrus) and vegetable from almost any country. For entry into the U.S., products must enter through specific ports and have a permit issued by APHIS Plant Protection and Quarantine. Additionally, the following requirements must be met:

- They must be imported from a definite area or district in the country of origin that is free from all injurious insects, or from certain specified injurious insects, that attack the fruit or vegetable, such that their importation can be considered to be without risk. If only certain specified insects are indicated, then all other injurious insects that attack the fruit or vegetable in the area or district of the country of origin must have been eliminated from the fruit or vegetable by treatment or other procedure(s);
- Surveys, adequate to detect any infestations, have been completed within the past 12 months by the plant protection service of the country of origin and have established the absence of infestations of injurious insects known to attack fruits and vegetables in the designated pest free area;
- Methods capable of preventing the introduction of injurious insects known to attack fruits and vegetables in the designated pest free area have been adopted and are being enforced by the country of origin; and

- The plant protection service of the country of origin has submitted detailed written procedures for the methods of survey and enforcement that will be used to prevent the introduction of injurious insects.

Once a country conforms to these requirements, regulations must be published in fulfillment of the Administrative Procedures Act before agricultural products are allowed entry into the U.S. without treatment subject only to inspection on arrival.

### **History of PFA**

The earliest efforts by USDA to recognize pest free areas began during the 1980s at a time when the U.S. Environmental Protection Agency initiated steps to cancel the use of ethylene dibromide, a fumigant widely used for quarantine treatment of fresh fruits and vegetables. In 1985, guidelines were issued to assist field managers in the initial assessment and subsequent development of proposals from foreign countries "to ship pest free agricultural products to the U.S. from export zones". This memo highlighted the importance of knowing the full complex of pests associated with the commodity, and indicated that the absence of biological information or approved survey methods may delay or prevent consideration of such requests. Work began immediately in a number of countries to develop workable proposals that were technically and operationally feasible.

USDA/APHIS published its requirements for pest free or definite areas in a final ruling on August 4, 1987. As of this date, four pest free areas have been officially approved by USDA/APHIS/PPQ. They are as follows:

**1. Chile.** The earliest and longest-standing recognition of a pest free area occurred in Chile in 1982. This program actually pre-dates the 1987 USDA regulations. Chilean efforts centered on an effective surveillance program for exotic fruit flies, a strong exclusion program, and immediate implementation and successful completion of

emergency procedures each time that adult Medflies were detected. The strong support and willingness to follow through on the necessary commitments initially provided by the Chilean private sector, and subsequently by the Chilean government have been significant factors toward the overall success of the PFA efforts in that country. This success gave tremendous impetus to formally establishing PFA requirements in the U.S. The Chilean government recently petitioned the USDA for official recognition of areas meeting the criteria of 319.56-2 (e) and (f) with regard to Mediterranean fruit fly which became effective on December 30, 1993.

*Approved areas:* All Provinces of Chile except Arica, Iquique and Parinacota.

*Approved commodities:* Apples; apricots; avocado; blueberry (fruit); cherry; Chilean cranberry (*Ugni molinae*); feijoa; kiwi (fruit); kumquat; loquat; Lucuma (*Manilkara sapota*); mango; mountain papaya (*Carica pubescens*); nectarine; *Opuntia* spp.; papaya; peach; pear; persimmon (fruit); plum; plumcot (fruit); *Pouteria* spp.; quince; *Ribes* spp.; rose hip; sand pear (*Pyrus pyrifolia*); sapote (fruit); and tuna (fruit). Cherimoya (fruit) and grape require an additional treatment due to the presence of other plant pests.

*Target pests:* *Ceratitis capitata* and *Anastrepha* spp.

**2. Sonora, Mexico.** Although fruit fly monitoring was conducted as early as 1981, efforts to establish a PFA formally began in 1985 with the establishment of fruit fly detection in both urban and commercial production areas. USDA and the Government of Mexico (GOM), through private grower support, provided personnel to service traps. Supervision and quality assurance monitoring were jointly conducted by GOM and USDA personnel. Apart from supervisory oversight, USDA and GOM also placed marked flies in traps as a quality assurance measure to verify that trap routes were properly serviced and trappers recognized the target pest.

The key to Sonora's success as a PFA lies in its geographic isolation from the closest infested areas of the country. The state of Sonora is bounded on the west by the ocean (Gulf of California) and on the east by the Sierra Madre mountain range. Vast arid desert areas surround each of the major production areas providing further protection from introduction or limiting the spread of new outbreaks.

Only two major highways enter the state which enables effective regulatory controls to be employed. An existing quarantine road station at Benjamin Hill, Sonora -- originally staffed only by USDA personnel as an added exclusion measure to protect to Arizona and California -- was relocated to Estacion Don, Sonora in 1987 to control vehicular traffic and commercial cargo entering the state from infested areas to the south. An additional road station was established on the highway entering the state in the north near Agua Prieta. The responsibility for staffing these road stations initially was shared equally by the U.S and by GOM. USDA actively monitors these activities, but the primary responsibility for surveillance and regulatory control now rests with GOM. Inspections of commercial vessels and trains are conducted by GOM near the towns of Guaymas and Empalme. Another road station located north of Guaymas provides an additional measure of quarantine security for those production areas to the north.

*Anastrepha ludens* continued to be detected within the state despite quarantine actions. Reservoirs of the pest persisted in populated areas that resulted from host fruits brought in from infested areas of Mexico. Fumigation chambers were installed at the road stations to treat host fruits which significantly lowered the incidence of outbreaks, particularly in commercial areas. Detections now were limited primarily to the vicinity of Magdalena in the northern portion of the state, and the town of Alamos in the southern reaches. Non-commercial (wild) hosts in these areas served as refugia for the pest. Efforts



focused on eliminating such hosts throughout the proposed free areas. Once these actions were implemented, outbreaks were further reduced.

GOM completed 12 months of detection trapping within 11 municipalities in 1987 and filed a written petition seeking formal approval of Sonora as a definite area. Final USDA approval for the PFA in Sonora was published in the Federal Register on March 29, 1988.

Since 1988, adults and larvae of Mexican fruit flies have been detected on several occasions within the approved PFAs. In each case, GOM, together with the Sonora Regional Agricultural Health Committee, has taken prompt action to delimit the outbreaks, implement the necessary control actions, and successfully eradicate the target pest before it has had a chance to spread.

*Approved Areas:* Eleven municipalities within the state of Sonora only.

*Approved Commodities:* Apple; apricot; grapefruit; sweet oranges; peach; persimmon; pomegranate; and tangerine.

*Target Pests:* *Ceratitidis capitata*; *Anastrepha ludens*; *A. serpentina*; *A. obliqua*; and *A. fraterculus*.

**3. Mossoro, Brazil.** Efforts to develop a PFA in Brazil began in 1985 with the development of a reliable detection method for the target pest. Prior to this date, very little scientific information existed on detection of the South American fruit fly, *Anastrepha grandis*. Methods, procedures and service intervals were adapted from trapping methods used for *Anastrepha ludens* and other related species. Trapping studies were conducted over several seasons in areas known to be infested with *A. grandis* and were compared to traps serviced in the area of Mossoro. Apart from demonstrating the absence of *A. grandis* from Mossoro, knowledge about other fruit flies of concern in the vicinity of Mossoro also was obtained and evaluated to determine if additional risk mitigation measures would be required.

Quarantine stations operated by the Government of Brazil are located on the principal roads to restrict the movement of commercial hosts into the fruit fly free area.

USDA periodically monitors the fruit fly surveillance and regulatory actions to verify the integrity of PFA status and insure that all necessary actions are performed in accordance with the annual work plan or protocol.

*Approved Area:* Mossoro area of Brazil (a portion of the state of Ceara).

*Approved Commodity:* Honeydew melons.

*Target pest:* *Anastrepha grandis* (only insect pest of quarantine concern to the U.S. known to attack honeydew melons).

**4. Riverland, Australia.** The Ministry of Agriculture, Forestry and Fisheries (MAFF) of Australia received PFA approval from the USDA in 1992 for citrus produced in certain locations within the Riverland district of South Australia. Trapping data was submitted in conformance with 319.56-2 (e) and (f) showing this area to be free from the Mediterranean fruit fly, Queensland fruit fly and other exotic fruit flies of U.S. concern. The presence of several citrus diseases in the area continues to restrict the movement of citrus from this area. MAFF is working to develop a systems approach to address these concerns.

*Approved Area:* 15 geographic subdivisions, called the "hundreds", of the Riverland district of South Australia, county of Hamley.

*Approved Commodities:* Oranges; lemons; limes; mandarins; and grapefruit.

*Target Pests:* Mediterranean fruit fly; Queensland fruit fly; and other exotic fruit flies.

## **Proposed PFAs Presently Under Consideration**

**1. Paraguana Peninsula, Venezuela.** Target Pest: *Anastrepha grandis*.

The Venezuelan Ministry of Agriculture and Livestock recently initiated surveillance efforts to

demonstrate that the melon fly, *Anastrepha grandis* does not exist in this region. Access to the peninsular region is extremely limited, so effective regulatory controls are possible. Past efforts have been hindered by a lack of coordination and commitment by the Ministry and private sector interests to follow through with the actions prescribed in the proposed work plan. Present efforts now appear to be better.

**2. Baja California Sur, Mexico.** Target Pests: *Ceratitis capitata*, *Anastrepha ludens*, *A. serpentina*, *A. obliqua*, and *A. fraterculus*.

The Secretariat of Agriculture, Livestock and Rural Development at the state and Federal levels have been working with local producers to eradicate *Anastrepha ludens* from Baja California Sur. Progress has been made in these efforts, but adequate regulatory controls do not yet exist to prevent reinfestation to the north of the quarantine road stations. Regulatory efforts could be improved by closer inspection of buses and installation of fumigation chambers. The uncontrolled entry of fruit fly infested host fruits from the interior of Mexico also must be controlled. The presence of *Anastrepha obliqua* in this state further complicates efforts to establish a PFA. Before a viable PFA can be established in this state, this species also must be eradicated.

**3. Chihuahua, Mexico.** Target Pests: *Ceratitis capitata*, *Anastrepha ludens*, *A. serpentina*, *A. obliqua*, and *A. fraterculus*.

This region contains many factors favorable to the establishment of a PFA: geographic isolation of commercial production areas by mountains and desert; climatic extremes that are unfavorable to fruit fly species; sufficient acreage and production of hosts; and a history of very low incidence of fruit flies. Road stations have been installed on the main highways at the southern extreme of the state. Improvements in fumigation procedures were recommended by USDA. Fruit fly surveillance measures are being carried out to demonstrate that the target species do not exist.

Once the trapping data has been compiled and submitted to USDA, the entire state of Chihuahua will be considered for PFA status.

**4. Provinces of Mendoza and Patagonia, Argentina.** Target Pests: *Ceratitis capitata*, *Anastrepha fraterculus* and other exotic fruit flies. Fruit fly eradication efforts are underway in these two provinces. Once eradication is completed, the Government of Argentina must actively continue to survey for a period of 12 months to demonstrate the absence of these two pests.

**5. Baja California Norte, Mexico.** Target Pest: Karnal Bunt.

A joint U.S.-Mexico technical working group is developing criteria for the survey of karnal bunt, an exotic disease of wheat. Once the survey methods are determined, GOM must actively continue to survey for a period of 12 months to demonstrate the absence of this disease. The CFRs must be amended to include plant diseases under the pest free area concept.

#### **Recognition of PFAs By Other Countries**

The following countries have established standards or other criteria for recognizing pest free areas for which they presently accept certification by the plant protection services of the exporting country or pre-clearance certification by inspectors from the importing country.

**New Zealand.** The National Agricultural Security Service (NASS), Ministry of Agriculture and Fisheries (MAFF) of New Zealand has published standards for fruit flies, citrus canker, and brown rot. Presently, MAFF has a bilateral quarantine agreement with Australia that permits the certification and entry of peppers from an area free of fruit flies. New Zealand also recognizes the U.S. as free from brown rot in accordance with the NASS standard for this disease.

**Australia.** In Queensland and the Northern Territory, the government has established growing periods during the months of May



through August when the Queensland fruit fly, *Bactrocera tryoni* cannot survive. During this period, all host fruits grown in these two provinces can be shipped to Victoria without treatment.

### **North American Plant Protection**

**Organization (NAPPO).** NAPPO became the first regional plant protection organization to formally adopt an international standard for pest free areas. This standard was approved by representatives of the three member nations -- Canada, Mexico and the United States -- on April 20, 1994. The standard provides the general requirements, definition of terms, and conditions for establishment, maintenance, termination and re-instatement. The standard also outlines general work plan requirements.

**Draft FAO Standard.** A draft proposal currently is being circulated by the Secretariat of the International Plant Protection Convention, Food and Agricultural Organization of the United Nations, among its member countries. The draft standard is entitled "Requirements for the Establishment of Pest Free Areas" Section 3 -- Surveillance of the International Standards for Phytosanitary Measures.

**North American Free Trade Agreement (NAFTA).** Provisions pertaining to pest free areas can be found in Section B: Sanitary and Phytosanitary Measures, Article 716: Adaptation to Regional Conditions.

**General Agreement on Tariffs and Trade (GATT).** Provisions pertaining to pest free areas can be found in Article 6 of the Agreement on the Application of Sanitary and Phytosanitary Measures of the Uruguay Round.

**Conditions and Situations Where Appropriate**  
Based on the knowledge and experience gained in the establishment and recognition of pest free areas approved thus far, efforts to establish or demonstrate an area to be pest free are greatly enhanced when the following conditions occur:

1. Natural geographic barriers such as an ocean or desert exist that exclude the target pest(s).
2. Isolation of growing areas from urban centers.
3. Specific commodities to be exported are poor or marginal hosts of the target pest(s).

Commodity treatments can sometimes adversely affect product quality. Damage can occur due to: 1) the treatment itself; 2) additional handling of the product; or 3) delay in its movement. This increases cost to the shipper and the consumer. Since pest free areas eliminate the need for commodity treatment, commodities that may be affected in this manner would be likely candidates for consideration of a PFA approach.

For a PFA to be operationally feasible, sensitive survey tools for detection of the target pests must be available. These detection devices or methods should be capable of detecting incipient infestations of a pest before it is able to spread beyond a radius of 1.5 miles and preferably before a new outbreak reaches a third generation. If the survey methods are not effective at low population levels, problems will be encountered in rapidly delimiting new outbreaks, thus making maintenance of the pest free area difficult.

Economic factors also are major determinants when considering establishing and maintaining a PFA. A sufficient level of commercial production of a commodity or commodities must exist to make cost recovery feasible. The high costs associated with the on-going requirements of

surveillance and regulatory measures must be supported entirely from contributions by the producers in the area or from fees assessed for inspections and treatments at quarantine road stations. Without adequate private sector support, PFA efforts will not succeed.

### **Opportunities and Limitations**

Existing regulations pertain only to injurious insects and would have to be amended to include plant pests and diseases other than injurious insects. Criteria for considering other plant pests and diseases are lacking and will require a great deal of time and effort to establish.

Due to the cyclic or episodic nature of some pests and diseases for which the biology and bionomics are poorly understood and/or for which inadequate methods of detection and control exist, pest free areas or regionalization may not be technically or operationally feasible. Pest reproduction and population dynamics will be different for each pest under different geographical and ecological situations. Pest levels may be statistically undetectable one moment, then explode in exponential proportions virtually overnight.

Fungi can grow for years without mating and producing sexual or asexual reproductive structures. However, once the reproductive trigger is met, a fungus can suddenly have great quantities of spores blowing in the wind. Such spores can be carried great distances by wind. Citrus black spot was extremely rare in South Africa for over 20 years, but suddenly became epidemic over large areas within a period of a few years.

Situations where multiple pest species or a complex of different pests occur may further limit efforts to establish pest free areas. In general, the risks associated with all target pests must be addressed or mitigated through other actions. Where technologies exist for detection or control of only one species within a pest complex, then it

becomes more difficult to establish and maintain an area pest free.

Before a PFA becomes operationally feasible, both surveillance and control technologies must be available and freely employed. Even though detection tools may exist, control technologies must exist to delimit, contain and eradicate all target pests of concern. If the necessary control technologies cannot be fully employed within the proposed PFA due to social, economic or environmental reasons, then the feasibility of establishing and maintaining the proposed area is questionable and may not warrant consideration.

Feasibility also depends, in large part, on the capability or competency of the plant protection services within a country or countries, and the degree to which such efforts are supported by both the private and public sectors.

Calibration or other data documenting the reliability of surveillance tools is frequently lacking. New trapping devices or methods should be compared with known detection systems to establish their overall effectiveness to detect the pest during times of low prevalence.

A common problem in many countries is the inability or unwillingness to regulate host commodities from nearby infested regions. For example, the populace of the city of Guayaquil, Ecuador depends heavily upon fresh host fruits and vegetables produced in other parts of the country. Although the Ecuadorian government conducts fruit fly surveillance in commercial production areas, the lack of regulatory controls on commercial host products entering this area prevents it from being recognized as a PFA by USDA.

Altitude alone, as a geographic barrier, could not be demonstrated in the case of *Anastrepha fraterculus* on *Rubus* species in Colombia. Although altitude may be a limiting factor due to the absence of certain hosts, temperature, etc.,



regulatory actions would still be required to prevent the introduction of infested host material into a PFA.

Pesticides are regularly employed to exclude target pests or eradicate outbreaks. Their use is subject to registration requirements. In some instances, use may be predicated on obtaining a special exemption, especially if the pest does not exist within a country. When this occurs, the operational feasibility of the proposed PFA may be questionable.

### Quantification of Risk/Biometrics

**Data Requirements.** As mentioned earlier, basic information on the biology of the target pest is extremely useful in assessing the technical and operational feasibility of a proposed PFA. The type of data that should be compiled and submitted along with a proposed work plan is the following:

#### *Life cycle and basic biology*

- number of generations per year - Multivoltine pests are more likely to become established in a new geographic area. It is easier to obtain data on such pests. Univoltine pests must be examined over a period of years, especially where diapause is involved by the pest.
- life stages present in or on commodity - All life stages may or may not be present in or on the commodity. To the degree that this information is known, it would be useful to evaluate risk associated with the commodity. All stages of the pest that are present must be addressed.
- susceptibility of life stages to detection and control
- level of infestation

- dispersal capability - An important factor in determining the size of buffer zones, if required, to maintain integrity of the PFA.

#### *Bionomics*

- habitat selection
- host/pest relationship
- maturity versus infestation rate
- alternate hosts

#### *Commodity*

- entire pest complex present

Also required are written details of the survey methods to be employed; type of trapping device; attractants that may be used; service interval for trap inspection; and quality assurance methods to demonstrate trapping effectiveness, including provisions for bioassay and chemical analysis of the attractants. Methods of detection also can include fruit collection/sampling and field surveys to demonstrate the absence of the target pests.

Details of exclusion measures also should include emergency response measures to be taken when an outbreak of the target pest occurs. A written protocol outlining emergency response measures should include: responsibilities; actions and timeframes; legal authorities to take necessary actions; increased surveillance plans to rapidly delimit the infestation; and confirmatory trapping plans to show that the pest has been successfully eradicated based on pest biology. Regulatory measures to be taken include: establishing quarantine boundaries; confiscation and proper disposal of infested fruits; and placing hold orders on premises to prevent movement within and outside of quarantine areas. Seizure/stop sale of host fruits in markets, fruit stands, and other safeguard measures may be appropriate.

The protocol also should identify the control measures to be taken along with implementation timeframes. The degree to which pesticides or other control measures are applied in a timely manner may be a critical factor in canceling only a portion of the PFA or the entire PFA.

Containment of the pest can be accomplished by fruit stripping the infested and surrounding properties. Applications should be based on biology of the pest with the aim of preventing reproduction.

Other work plan requirements are cited in the NAPPO Standard for Pest Free Areas dated April 20, 1994.

### **Marketing Practices**

Marketing practices have not been a consideration in approval of PFA by USDA except from the standpoint of safeguarding procedures to prevent possible infestation and methods/routes of transportation. Such practices have had an important bearing on the economic viability of PFAs particularly as they pertain to the volume of host fruit and seasonal aspects of marketing.

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## Systems Approaches

### Introduction

A systems approach is a set of safeguards and  
mitigation measures which individually and  
cumulatively provide a reduction in plant pest  
risk. This approach to management of plant pest  
risk provides an alternative to post-harvest  
quarantine treatment for obtaining phytosanitary

security. The safeguards and mitigation measures  
can occur in the growing area, at the  
packinghouse, or during the shipment and  
distribution of the commodity. Post-harvest  
treatments can be components of a systems  
approach. The components of a systems approach  
can vary widely, but commonly include pest  
survey, trapping and sampling, field treatment,  
cultural practices, host resistance, post-harvest  
safeguards, limited harvest period, limited sales  
distribution, and restrictions on maturity at  
harvest.

In general, a systems approach has two or more  
components that are independent, thus creating  
redundancy. That is, it operates like a fail-safe  
system in that if one mitigating measure fails,  
other safeguards exist which still reduce the risk  
to a negligible level. An example of this is the  
application of a systems approach to achieve a  
pest free glasshouse. The selection of both screen  
vents and double doors does not provide  
redundant or independent safeguards. If one fails,  
the pest can enter the glasshouse even though the  
other safeguard functions perfectly. An  
independent measure for this example would be  
pest survey (trapping) in the glasshouse environs.  
Now, either the pest survey or the glasshouse  
exclusion safeguards could fail, but if the other  
component works, the host plant within the  
glasshouse remains uninfested.

Not being considered as systems approaches in  
this document are: 1) combination treatments,  
where two post-harvest treatments are used to  
reach the required mortality; and 2) situations  
where post-treatment safeguards are required to  
eliminate the risk of reinfestation. These are  
included in the Probit 9 section. An approach that  
is being considered as a systems approach  
involves inspection to ensure a low infestation  
rate, combined with a post-harvest treatment.

### Current Applications

**A. General.** The United States Department of  
Agriculture (USDA) has used systems approaches

to allow the safe movement of agricultural commodities for many years. These include both exported and imported products, as well as commodities transported from one area in the United States to other U.S. locations. Examples of current successful applications of the systems approach are discussed in this section.

**B. Unshu oranges from Japan.** Generally, USDA prohibits the entry of fresh citrus fruit from locations where citrus canker exists. For about the last twenty years, Unshu oranges have been permitted entry into the United States with a systems approach. This systems approach requires an established growing area with surrounding buffer zones, field survey, surface treatment, resistant varieties, inspection, and limited U.S. distribution.

**C. Citrus Fruit from Florida.** The Caribbean fruit fly, an introduced pest in Florida, is not considered an economic pest of citrus, but does sometimes attack citrus fruit. Oranges, grapefruit and other citrus fruit are certified for movement from Florida to other citrus growing states and foreign locations, including Japan, by the use of a systems approach. The components of the Caribbean fruit fly program include poor-host status, removal of preferred host, established growing area with buffers, trapping, field treatment, restricted harvest periods and fruit cutting.

**D. Bell Peppers from Israeli.** Generally, USDA prohibits the entry of fresh peppers into the United States to prevent the introduction of the Mediterranean fruit fly from areas where this pest exists. Mediterranean fruit fly occurs in Israel but peppers are allowed entry under a systems approach. The requirements for this approach include growing the host within a fly-proof glasshouse, location of the glasshouse in areas where the pest is absent or rare, trapping of the environs, and fly-proof packaging.

**E. Citrus fruit from Texas.** The Mexican fruit

fly occurs in southern Texas and is a serious pest of citrus fruit. A systems approach is used instead of a post-harvest treatment for the certification of orange and grapefruit from the lower Rio Grande Valley. This allows for unrestricted interstate movement of the fruit and for export to certain countries. The components of this program include certification for areas in three Texas counties, release of sterile flies, trapping, and malathion bait treatment.

### Requirements

In general, systems approaches are more difficult to manage than a probit 9 post-harvest treatment or a pest free area. Many of the components need to be supervised or monitored to ensure compliance. For certain pest/host combinations, the population level of the pest, the biology of the pest, or the growing requirements of the host make the development of a systems approach impractical. Listed below are conditions and situations that allow for a successful system to be developed.

- Pests associated with the commodity are known.
- Basic biology of the pest(s) is known, including pest/host relationship, dispersal, alternate hosts, habitat selection, and population dynamics.
- Systems exist for field surveillance and/or detection of pests in shipment.
- Knowledge of harvesting, packing, and marketing practices.
- Pest(s) generally absent or rare in commercial commodity because of : 1) normal field management; 2) poor host; 3) resistant cultivar; 4) phenological asynchrony between pest and commodity; or 5) ecological limitation-based rarity of pest in growing area..)



- No alternative method is available for obtaining phytosanitary security; or a systems approach is more desirable because it does not damage the commodity and/or is more cost-effective.
- Sufficient volume of the commodity is shipped to justify and offset the program costs.
- Some degree of redundancy and independence between program components can be designed to allow for variability in pest populations or partial failure of other components.
- Phytosanitary security is apparent either by qualitative or quantitative assessment.

### Opportunities and Limitations

As chemically based post-harvest treatments become less available, more reliance will be placed on alternative methods, including systems approaches. Because systems approaches require at least two components, and generally several, the management of this method is difficult and the cost of the programs can be high. Since systems approaches rely heavily on a sound knowledge of the pest and host biology and how they relate to each other, the programs can also be costly and time-consuming to develop. The pest-host relationship of some pests and their preferred hosts can make the development of a systems approach unfeasible.

Even with the limitations listed above, the desire to allow the movement of currently prohibited products or the need to find an approach that does not damage the product will encourage the development of new systems approaches. Current improvements in risk assessment being implemented by USDA, which will better quantify the overall risk or level of security, will promote a wider acceptance of the systems approach by U.S. industry, state cooperators, and trading partners.

### Assessment of Risk

**General Comments.** Risk assessments conducted to support decisions concerning trade in agricultural commodities have historically been qualitative. This is certainly true in situations where plant pest risks have been managed with a systems approach. Each of the examples of systems approaches discussed earlier in this chapter relied on qualitative risk assessments. Use of certain quantitative methods (see below) is relatively new. In many cases, the additional effort needed to conduct a quantitative assessment is not justified because of limited risk or limited commercial interest in the commodity. Both qualitative and quantitative risk assessments can lead to good, well-informed decisions.

Phytosanitary treatments, such as methyl bromide fumigation at a rate sufficient to kill 99.9968% of treated individuals (*i.e.*, Probit 9 mortality), provide a familiar framework within which to consider mitigation of plant pest risk. However, using a phytosanitary treatment with proven efficacy of probit 9 mortality is not a risk-based standard. While such a treatment can be expected to reduce the frequency of pests by a predictable amount, the risk of pest introduction remains a function of the infestation rate of the treated commodity.

When a systems approach that does not include a phytosanitary treatment is used to manage plant pest risk, the familiar framework of a phytosanitary treatment with efficacy of probit 9 mortality is lost and alternative methods for assessing risks are needed. As described above, systems approaches involve two or more components. The diversity of components presents the problem of trying to express in similar terms the degree to which the various components affect plant pest risk.

The efficacy of various mitigation measures can seldom be expressed in common terms, and seldom in the same terms as a treatment. For

example, a probit 9 treatment provides a demonstrated level of mortality (*e.g.*, 99.9968% mortality of treated individuals); pesticide treatments in the growing area reduce the incidence of pests as evidenced by reduced trap captures; and culling at the packinghouse removes 100% of the pests that are detected, but only some of the pests that may be present in the commodity. In addition, some components of a systems approach are not mitigation measures at all, but a means to assess the presence or absence of pests (*e.g.*, trapping surveys, biometrically-based inspections). Clearly, with a systems approach, more detailed methods for quantifying risk are needed.

Regardless of whether a qualitative or quantitative method is used, at least the following should be considered:

- identification of pests associated with the plant species
- pests which are of quarantine significance
- pertinent information on significant quarantine pests
- consequences of introduction of the quarantine pest
- likelihood of introduction of the quarantine pest

The other important part of an overall pest risk analysis is the risk management phase which should include consideration of at least the following:

- identification of appropriate mitigation measures
- judgement concerning which mitigation measures are optimal for managing the risk

- judgement concerning whether the risks can be mitigated sufficiently
- pest risk mitigation options
- implementation and monitoring of any mitigation plan adopted

### Quantitative Risk Assessment

In order to express the overall efficacy of the systems approach, a "common currency" (*i.e.*, term of expression) is needed when estimating plant pest risk. The chosen endpoint for the risk assessment affects how risk can be expressed throughout the assessment. Examples of risk assessment endpoints include:

- Frequency of entry<sup>3</sup> (*i.e.*, number of pests entering per unit time).
- Probability of entry (*e.g.*, probability of pest entry per unit of commodity imported).
- Frequency or probability of establishment.
- Frequency or probability of pest outbreaks.

Regardless of endpoint, the risk assessment should be consistent with the endpoint.

Scenario analysis is an established risk assessment method that has recently become more common in pest risk assessments. Scenario analyses have been used to characterize pest risks in the U.S. (*e.g.*, Griffin and Miller 1995), Canada (*e.g.*,

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<sup>3</sup> The maximum pest limit concept (Baker et al. 1990) which was designed originally for fruit flies has two primary components: a partial risk assessment (*i.e.*, the likelihood of introduction component of a risk assessment) and a pest risk management decision standard (*i.e.*, "...the maximum number of immature fruit flies that may be present in consignments imported during a specified time to a specified location"). A maximum pest limit of three live larvae per day is a pest risk management decision standard.



Renwick 1994) and Australia (e.g., Phillips et al. 1994). Additionally Monte Carlo simulation methods have been used to estimate the probability/frequency of pest introductions in the U.S. (e.g., Griffin and Miller 1995) and Canada (e.g., Renwick 1994) among others.

**Example: Importation into the U.S. of Mexican avocado fruit.** In 1995, APHIS conducted a pest risk assessment (USDA 1995) following a request from the Mexican government to allow importation of Mexican avocado fruit. The endpoint chosen for the risk assessment was the frequency of U.S. outbreaks of exotic pests from Mexico as a result of importations of Mexican avocado fruit. The quantitative portion of the risk assessment to estimate the frequency of pest outbreaks consisted of:

**Scenario Analysis.** The scenario analysis identified a series of independent events (nodes) comprising the scenario leading to a pest outbreak (e.g., the commodity must be infested; the pest is not detected during harvest and packing; the pest must survive shipment; the pest must find suitable host material; etc.). The scenario analysis provides a conceptual framework within which to estimate the frequency of pest outbreaks.

**Development of a mathematical model.** In order to express the endpoint (frequency of pest outbreaks, a quantitative parameter), an equation (i.e., mathematical model) was developed to calculate the estimated frequency of outbreak for each pest. The mathematical model was based on the scenario analysis. Because of the way the scenario analysis was constructed (i.e., a series of independent events that must all occur to result in a pest outbreak), the appropriate model was a simple, linear, multiplicative model.

The first component of the model was an estimate of the frequency of importations of avocado fruit. This estimate was then multiplied by a series of probabilities, one estimate for each independent event. Because the frequency of importations was

expressed in terms of the number of 16-fruit boxes imported per year, and all probability estimates were estimated on a per box basis, the model provided output in terms of pest outbreaks per year.

**Estimation of probabilities.** Probabilities were estimated for each node (event) identified in the scenario analysis. Instead of relying on point-estimates for probabilities, a frequency distribution of possible probability values was specified for each event, for each pest. This is an explicit and transparent way to account for the uncertainty involved in estimating probabilities for events for which the true probability can never be known.

**Monte Carlo calculations.** It would have been possible to use a single probability estimate for each of the nodes identified in the scenario analysis and to estimate the frequency of pest outbreaks based on a single calculation. Monte Carlo calculations account for the uncertainty in the probability estimates for each node by calculating the estimated frequency repeatedly. For each pest, for each of two program alternatives, the estimated frequency was calculated 1,000 times. For each of the 1,000 calculations, the computer program randomly selected a value for the frequency of importations and each component probability from the specified probability distribution. Thus, the output of the model provided a probability distribution of estimates for the frequency of pest outbreaks for each pest, for each program alternative. The output was expressed in terms of the frequency of pest outbreaks per year and the number of years between pest outbreaks.

## SECTION II. Potential Strategies

### Alternative Treatment Efficacy

#### Introduction

The use of probit 9, i.e., 99.9968% mortality or 32 survivors per million treated population, as the acceptable level of quarantine security was recommended over five decades ago (Baker 1939), and has since been the practiced dogma. With mortality of the treated population as the criterion, probit 9 provides adequate quarantine security for heavily infested commodity. However, treatment based on probit 9 requirement may be too severe for commodities that are rarely and poorly infested. For instance, assuming a shipment of 36,000 fruit and a 1% infestation (one larva per fruit), a disinfestation treatment with probit 9 mortality (or 0.0032% survival rate) will have an expected number of  $36,000 \times 0.01 \times 0.000032 = 0.012$  survivors. In this case, probit 9 is too stringent a requirement. For the same shipment size and infestation rate, the survival rate ( $s$ ) that can be expected to produce one survivor can be calculated:  $36,000 \times 0.01 \times s = 1$ , with  $s = 1/360 = 0.0028$  or 0.28%. Thus, a treatment with only 99.72% mortality or probit 7.77 is needed. A less severe, lower probit treatment is deductively more economical, in addition to better preserving the quality of the commodity.

The probit 9 standard is practiced independent of: rates of infestation; gregariousness (single versus multiple infestation); survival and reproductive capacity of the pest; inherent hardiness of the pest to environmental stress during packaging and shipment; packaging and shipping conditions; seasonality of shipment; distribution of commodity; and other biological and nonbiological parameters. In situations where the natural field infestation rate is low and the post-harvest survival and reproductive capacity of the pest are inherently poor, the application of probit 9 standard may be too rigid, impractical, and

unnecessary (Landolt et al. 1984, Moffitt 1989, Baker et al. 1990, Vail et al. 1993). Following this argument, an alternative treatment efficacy (i.e., treatment efficacy other than probit 9) approach in combination with modifications in inspection and marketing strategies could attain the required quarantine security. In the alternative treatment efficacy approach, the risk is measured as the probability of survival of one or more mating pairs in a shipment (Landolt et al. 1984, Vail et al. 1993). In addition to a mating pair, a reproductive unit may also be a parthenogenic individual or a gravid female.

#### Current Policies

Protocols that estimate the required treatment efficacy based on the probability of introduction of a mating pair or a reproductive unit are evolving and have not been adopted.

#### Requirements

The main quantitative criterion for deviating from the probit 9 regulatory standard is a naturally low level of pest infestation in the commodities to be marketed. If this low infestation rate is a manifestation of inherently low survival and reproduction of the pest, then the effect of various biological and ecological factors on the life history parameters of the pest must be documented, with special emphasis on the life history of the pest on the commodity being considered for export. However, if the low infestation rate is a manifestation of the relative resistance or a poor host status of the commodity, then extensive field surveys are needed to document variation in infestation level due to variety/cultivar of the host, condition of the fruit, seasonality of production, and biogeography of the production areas. This data should be supplemented with forced laboratory infestation studies and a review of historical, worldwide data on the host plant/pest relationship to facilitate the determination of host status and carrying capacity of the host. It is important to realize that one host plant species may be a poor host or non-host at one location, but a preferred host or routinely



utilized host at another location. In some cases, infestation data may have to be specific to the cultivar and production fields or orchards.

Other factors that may be considered in this approach include the frequency distribution of number of pests in the fruit; determination of other pests which attack the commodity; other host plants of the pest; shipment size; seasonality of shipment volume; shipping and storage conditions; use and distribution of the commodity; and culling practices used to remove pest-damaged fruits.

### Opportunities and Limitations

Post-harvest quarantine treatments that have been designed following the requirements of the probit 9 standard mortality inflict severe damage on many commodities, rendering them non-marketable. In these commodities (mostly climacteric, tropical species), the opportunities that the alternative treatment efficacy approach might bring are tremendous. If the commodity is a marginal or poor host and can tolerate a treatment with requirements of mortality lower than probit 9, then it may be possible to export the commodity without causing any significant threat of pest introduction. This approach, however, may be applicable only to commodities with low infestation rates.

### Quantification of Risk

Here, the main criterion for defining risk is the probability of a potential mating pair surviving the quarantine procedures. The probability of finding at least one or more mating pairs is given by the equation (Landolt et al. 1984):

$$P = \sum_{x=2}^{x=\infty} (e^{-NR} (NR)^x / x!) * (1 - 0.5^{x-1}) \quad (1)$$

where  $P$  is the probability of having at least one or more mating pairs per shipment;  $N$  is the number of fruit or kg of fruit with  $R$  (average number of larvae per fruit) infestation rate; and  $x$

is the number of larvae in the shipment, assuming a Poisson distribution of the larvae infesting the commodity. Appendix A presents a program written in SAS language which can be used to calculate  $P$  based on Equation (1) (SAS Institute 1995). Equation (1) can be simplified as (Baker et al. 1990) :

$$P = [1 - e^{-NR/2}]^2 \quad (2)$$

Based on equation 2,  $NR$  as a function of  $P$  can be calculated as (Vail et al. 1993):

$$NR = -2 * (\log_e(1 - \sqrt{P})) \quad (3)$$

where  $P$  is the probability of one or more mating pairs and  $1 - P$  is the probability of <1 mating pair being present. Knowing  $NR$ , the level of mortality ( $m$ ) that will be required from a disinfestation treatment can be calculated, given the following information: infestation rate ( $i$ ), expressed as number of larvae per kg of host product; the quantity of the commodity to be shipped ( $n$ ), in kg; and the natural survival rate ( $s$ ) in the host for export. The equation to calculate the level of the required commodity quarantine treatment mortality ( $m$ ) is:

$$m = 1 - (NR / (i * n * s)) \quad (4)$$

where  $NR$  is the total number of pest allowed as a function of  $P$ . Vail et al. (1993) presented excellent examples on the calculation of  $NR$  and  $m$  for various host plants infested by codling moth.

The required  $m$  will vary with commodity and pest. Adjustments on the required  $m$  can be made based on: the expected infestation levels at harvest and during packaging and shipment; the shipment size; the movement of the commodity in

the importing country; the pest's behavioral attributes to find food and mate (e.g., lekking); and other factors that may influence the calculations of risk to provide quarantine security while preventing unnecessary treatment overkill. Appendix A presents a program written in SAS language which can be used to calculate  $m$ , based on Equations (3) and (4).

Another approach to the quantification of risk is to express equation (2) as a function of field infestation rate and the expected survival rate of the pest after the disinfestation treatment. Equation (2) can be expressed as:

$$P = [1 - e^{-NFT/2}]^2 \quad (5)$$

where  $F$  is the field infestation rate and  $T$  is pest survival after the disinfestation treatment. Therefore,  $NFT$  would be the average number of live pests in a shipment after a post-harvest treatment.

When the male to female ratio is not 1:1, equation (2) can be written to calculate  $P$  as:

$$P = [1 - e^{-NFAx}] * [1 - e^{-NFB y}] \quad (6)$$

where  $A$  is the proportion that are males;  $B$  is the proportion that are females;  $x$  is the survival rate for males after treatment; and  $y$  is the survival rate for females after treatment.

In situations where the reproductive unit is not a mating pair, i.e., parthenogenic individual or gravid female, the probability of one or more reproducing individuals, assuming a Poisson distribution, can be expressed as:

$$P = 1 - e^{-NRT} \quad (7)$$

In applying equation (7) to assess pest risk for population with both sexes in which females are

assumed to be gravid upon arrival at commodity destination, only the number of females needs to be determined to estimate  $R$ .

### Examples

Table 1 gives self-explanatory examples of estimating probability of one or more reproductive units ( $P$ ) and the required level of commodity treatment efficacy ( $m$ ) for certain commodities. In examples 1, 2, and 3, the probability of one or more mating pairs are calculated for: Caribbean fruit flies in guava; oriental fruit flies in mature green to half ripe 'Kapoho' and 'Sunrise' varieties of papaya; and oriental fruit flies in mature green to half ripe 'Arkin', 'Kajang', 'Kari', and 'Sri Kembangan' varieties of carambola. Based on these commodities with approved treatments that adhere to the probit 9 requirements, the probability of having one or more mating pairs ranges from 0.00031 (oriental fruit fly in 'Sunrise' papaya that are shipped by air) to 0.0022 (Caribbean fruit fly in guava shipped by sea).

In example 4, rambutan, a known poor host of oriental fruit fly, is given a treatment with probit 8.1 efficacy (99.9% mortality). The resulting  $P$  values are comparable to those of examples 1, 2, and 3 that received treatments with probit 9 efficacy.

Oriental fruit fly infestations in examples 5 and 6 are from harvest mature 'Bosworth', 'Brewster', 'Groff', 'Kaimana', 'Kwai Mi', and 'Tai Tso' cultivars of lychee. In here, the calculation of required treatment efficacy is illustrated, given the values of infestation rate, survival rate, and the size of shipment. Based on the workshop participants assessment, the value of  $P$  could be set at  $\leq 0.01$  for pests that are high risks to U.S. agriculture, and a value of  $\leq 0.05$  for pests that are low risks. In these examples, the calculated  $m$  are much lower than probit 9.



Example 7 is hypothetical: fruits are infested at a rate of 0.1 pest per fruit; 2 males:3 females; and, the only known treatment that preserves fruit quality has an efficacy of 99.9903% mortality (probit 8.72) for males and 99.9% mortality (Probit 8.1) for females. Using equation (6), the probability of at least one mating pair being in a shipment is 0.0522.

Example 8, also a hypothetical example, shows the probability estimate of one or more reproductive units for a pest that does not require a mate, either a parthenogenic individual or a gravid female.

Appendix C is a software package called Commodity Quarantine Treatment Statistics (CQT\_STATS). This is an IBM-PC based specialty calculator that provides user-friendly computation of Equations (1) thru (7).

## **Summary**

The argument for alternative treatment efficacy approach is that the probit 9 requirement is too severe for many commodities that are poor hosts of the quarantined pest. A less severe treatment with efficacy level lower than probit 9, when combined with modifications in packing, distribution, and inspection, may attain the desired quarantine security. The alternative treatment efficacy approach estimates risk of pest survival and establishment based on biological, ecological, quarantine treatment, and marketing or distribution data. Risk is defined as the probability of one or more reproductive units being present in a shipment. A reproductive unit may be a mating pair, a gravid female, or a parthenogenic individual. Further definition of risk that should be addressed is the probability of the mating pair to mate, reproduce, and find suitable host in the area of commodity destination. For gregarious pests, an interesting question would be the probability of a mating pair being present in the same infested commodity.

Table 1. Examples showing potential applications of the concept of alternative treatment efficacy.

Example No. Commodity	Variety	Pest	Shipment		Infestation rate/fruit	Survival rate	Treatment efficacy		Probability of at least one reproductive unit	Equation
			type	size			%	Probit		
1. Guava		Caribbean fruit fly	sea	1,000,000	0.003		99.9968	9	0.0022	5
2. Papaya	'Kapoho'	Oriental fruit fly	sea	11,900	0.3994		99.9968	9	0.005362	5
			air	3,500	0.3994		99.9968	9	0.000489	5
	'Sunrise'	Oriental fruit fly	sea	11,900	0.1004		99.9968	9	0.000358	5
			air	3,500	0.1004		99.9968	9	0.000031	5
3. Carambola		Oriental fruit fly	air	50,000	0.0906		99.9968	9	0.00489	5
4. Rambutan		Oriental fruit fly	sea	268,000	0.0004		99.9	8.1	0.002724	5
			air	33,600	0.0004		99.9	8.1	0.000045	5
5. Lychee	'Bosworth' 'Brewster' 'Groff' 'Kaimana' 'Tai tso' All varieties	Oriental fruit fly	sea	20,000	0.0005	0.3054	93.1001	6.48	0.01	4
				20,000	0.0040	0.3054	99.1375	7.38	0.01	4
				20,000	0.0053	0.6250	99.6819	7.72	0.01	4
				20,000	0.0065	0.2446	99.3373	7.47	0.01	4
				20,000	0.0017	0.2946	97.8962	7.03	0.01	4
				20,000	0.0038	0.3550	99.2189	7.41	0.01	4
				20,000	0.0003	0.4268	91.7713	6.38	0.01	4
6. Lychee	'Groff' 'Kaimana' All varieties	Mediterranean fruit fly	sea	20,000	0.0003	0.1518	76.8642	5.73	0.01	4
				20,000	0.0001	0.2893	63.5808	5.34	0.01	4
7. Hypothetical example:	$\sigma^7$ : $\varphi$ ratio is 2:3; susceptibility to treatment differs by sex		sea	20,000	0.1		99.9903 $\sigma^7$	8.72 $\rangle$	0.0522	6
							99.9 $\varphi$	8.10 $\rangle$		
8. Hypothetical example:		gravid female	sea	20,000	0.1		99.9968	9	0.0620	7



## Maximum Pest Limits

### Introduction

The maximum pest limit concept refers to a method for estimating the intensity of inspection or mitigation necessary to provide a given level of security against pest introduction based on the probability that a founding population would be present in a designated lot.

For commodities that are considered preferred hosts, and in cases that a founding population would frequently be found in a single shipment or single commodity, the technique may provide a means to define inspection intensity when pests are detectable. Current and traditional methods focus on the probability of pest presence, but the maximum pest limit concept is specifically designed to consider the probability of establishment.

In the case of rarely infested or very poor hosts, the method provides a means to more precisely estimate the degree of efficacy required to achieve quarantine security. These conditions would include the use of historical data demonstrating rare infestation or low carrying capacity of specific commodities.

Four related publications have addressed practical ways to estimate risk of pest survival from ecological and treatment data. Landolt et al. (1984) proposed that the probit 9 mortality level for disinfestation treatment was excessive when potential hosts are not attractive to the pest of concern (therefore, trapping rate or commodity infestation rate is low), when survival in a potential host is low, or when pest population densities are low. In these cases, the vast majority of individual commodities have no pests, and the pests' distribution among infested commodities would be approximately a Poisson distribution. Infestation rates and risks of Florida citrus becoming a vector for the spread of the Caribbean fruit fly was given as an illustration of "overkill" resulting from requirements for probit 9 (=

99.9968%) mortality levels.

Couey and Chew (1986) presented a method for estimating the number of pests that must be treated with 0, 1, 2...survivors in order to demonstrate, at a given upper confidence level, a given treatment efficacy (such as  $\leq 0.000032$  survival). Their methods are frequently used in postharvest treatment research to determine the sample sizes for confirmatory tests. These calculations determine the numbers of pests that should be treated with maximum survival rate in order to estimate a confidence interval for a given efficacy; e.g. >92,000 larvae killed with no survivors in order to have 95% confidence that treatment efficacy is  $\geq$  probit 9.

Baker et al. (1990) published a method to estimate the maximum permissible rate of fruit infestation (i.e., a certain number of survivors entering a location), given known numbers of pests per fruit, a known post-harvest treatment efficacy, and known numbers of fruit entering a location during a period when reproductive stages of the pests could encounter each other. The Baker et al. (1990) calculation does not require a Poisson distribution of pests among commodity units (fruits for this discussion). However, following treatments, the calculation method assumes a Poisson distribution of survivors among fruit. Vail et al. (1993) used similar methods, given a known survival rate and number of fruit, to calculate the probability that a mating pair would survive a shipment.

### Current Policies

Protocols that use a computation of a maximum infestation percentage, a maximum pest limit, or that calculate sample sizes for commodity inspection based on the methods described in these papers have not been adopted in practice by regulatory agencies. However, USDA-APHIS has codified a few inspection programs based on a maximum infestation threshold using a hypergeometric sampling scheme. Preclearance inspection programs for apples from Europe,

Australia, and New Zealand, and an inspection program for stonefruit from Chile are examples of this application.

### Conditions and Situations where Appropriate

The major requirement for the application of any of these methods is a set of reliable estimates for infestation. Treatment efficacy data are usually available as a dose-response function in published literature for any approved quarantine treatment. Well known quarantine insects have extensive published surveys of infestation rates but these are highly dependent on commodity, growing conditions, and pest management practices in the production area.

The approach used by Baker et al. (1990) and Vail et al. (1993) estimates the carrying capacity of the host for an individual insect. If the carrying capacity (the maximum number of reproductive pests that can be produced independent of the number of eggs laid on commodity) is not affected by local ecological factors, this number is likely to be reliable; e.g., 1 or 2 pests per fruit for codling moth on cherries, or 40 pests per mango for Mexican fruit fly.

Vail et al. (1993) use  $[(N) \times (R)]$  as the total number of commodity units (fruit) = (N) multiplied by the infestation rate (R) to estimate (P) the probability of a surviving pair.

$$P = [1 - e^{-NR/2}]^2 \quad (2)$$

This equation can easily estimate the risk of a pair being introduced if: 1) the number of fruit entering a location is known; 2) the rate of infestation is known; and 3) the infestation rate follows a Poisson distribution. The calculations required can be done on any calculator that can calculate  $e^x$ .

The Baker et al. (1990) technique attempts to account for the non-Poisson distribution of pests among commodities (clumped for most fruit flies)

by separating the components of (R) into mean number of larvae per infested fruit ( $=\mu$ ) and treatment efficacy ( $=\phi$ ) so that survivors per fruit ( $=\lambda$ ) is  $\mu\phi$ , the product of larvae per fruit multiplied by the treatment efficacy. With the assumption of high treatment efficacy, Baker et al. (1990) then assume that the distribution of surviving larvae among fruit ( $=\lambda$ ) follows a Poisson distribution.

### Quantification of Risk

The maximum proportion of infested fruit (p = number of fruit with at least 1 pest/total fruit) in a total lot of (N) fruit can be estimated with a confidence by solving the following equation:

$$\sum_{r=0}^m \frac{(\mu \phi N p)^r}{r!} \exp(-\mu \phi N p) - \sqrt{\alpha} = 0 \quad (8)$$

By letting  $\mu\phi N = x$ , the equation can be solved for (p) in *Mathematica* (Wolfram Research, Inc. 1993) as :

FindRoot[(E<sup>-x\*p</sup>)\*Sum[((x\*p)<sup>r</sup>/r!, {r, 0, mpl} ])-(0.95<sup>.5</sup>)==0, {p, .01}, MaxIterations->60].

The value (x) is the expected number of larvae surviving in a lot (after treatment) and is the same as  $[(N) \times (R)]$  in the Vail et al. (1993) equation. The real difference between the two papers is that Baker et. al. (1990) allow separate entries for treatment efficacy and the number of larvae per fruit, so that (p), the maximum proportion of fruit infested, can be estimated.

If the number of larvae per fruit has a highly clumped distribution (as is known for many fruit flies), then the number of larvae per infested fruit will be relatively high, and the number of uninfested fruit will be low. If the treatment mortality is high, (>99%), then clustering of larvae will only be important if the number of survivors is >1 for an individual fruit.



The Baker et al. (1990) paper cites Couey and Chew (1986) in the references but does not give the equations used to determine the number of fruit that must be inspected (usually by destructive cutting) with no pests (dead or alive) per lot in order to demonstrate that (p) is below a certain level. Calculations with actual data for fruit cut indicate that the required number estimated with confidence "C" in Couey and Chew formulae are not the same as those estimated in Baker et al. (1990).

The calculation method of Baker et al. (1990) apparently used the equations similar to those in Couey and Chew (1986), but assigned an  $\alpha$  value of 0.95, and used  $(1-\sqrt{\alpha})$  in the equations to estimate (mx) (the number of fruit to inspect to demonstrate that the true infestation rate is not higher than p):

$$\exp(-p*mx) = (1-\sqrt{\alpha})$$

This can be solved in *Mathematica* with p (the maximum allowable proportion of infested fruit) as:

```
frcut=FindRoot[(1-( $\alpha$ ^0.5))-(E^(-p*mx))=0,
               {mx, Random[]}, MaxIterations->60]
```

Table 2 lists the results from Baker et al. (1990) and from calculations using *Mathematica* and formulae from Couey and Chew (1986). Those results were nearly identical for the values of "p", but were different for the calculation of the estimate for "n". When the value of  $\alpha$  is checked by using the Baker et al. (1990) values for n and p, most of the differences could be accounted for as rounding errors. When p is rounded at the  $10^{-3}$  level (as Baker et al. 1990 did in the publication), rather large ( $\pm 1\%$ ) errors result when the "n" value is calculated.

The use of  $\sqrt{\alpha}$  in Baker et al. (1990) rather than  $\alpha$  in the equation is curious. Correspondence with Robert Mangan from the New Zealand group did not offer an explanation for this, but the value  $\sqrt{\alpha}$

could have been designed to approximate a two-tailed test. Couey and Chew (1986) clearly state their justification for the one-tailed test. All that can be deduced is that  $1-\sqrt{\alpha}$  in the Baker et al. (1990) calculations corresponds to C values of 0.9747 in Couey and Chew (1986), and that the numbers of fruit cut for the demonstration data herein match those in Couey and Chew (1986) for  $s = 0$  by using  $(1-C)/2$  in place of  $(1-\sqrt{\alpha})$ . This would give (for two-tailed test)  $C = 0.95$ ,  $(1-C)/2 = 0.025$ , which in the program predicts a need to cut 899.9 fruit. Using  $C = 0.975$  in Couey and Chew (1986) with  $s = 0$  results in 900 fruit cut. As shown in Table 2, if the estimate of n is made using  $C = 0.95$  rather than  $C = 0.975$ , the values for n are reduced considerably.

Table 3 illustrates calculations of probability of mating pair and maximum pest limit using field data for oranges and mangoes from fruit production regions of Mexico. Calculations were made using the Vail et al. (1993) (probability of a mating pair) and the Baker et al. (1990) (maximum pest limit) equations to determine the degree of security provided by the probit 9 treatment level.

Four sample sets were used (see Table 3): The "oranges on ground" and "mangoes on ground" sets were collected at two sites in Baja California Sur (Candelaria [C] and San Jose Viejo [SJV]) in 1992 and 1993. A Mexican fruit fly eradication program was initiated in 1993 at San Jose Viejo, and Candelaria was treated with sterile flies in 1992 and 1993. The "oranges-on-tree-no treatment" and the "oranges-on-tree-with treatment" sets were collected in the LLera region of Nuevo Leon (NL) in 1991. This was an untreated orchard near alternate hosts for Mexican fruit fly. The season data included samples picked from trees at four (early, mid, late, end) intervals during the season with no treatment. The "oranges-on-tree-with treatment" set had samples that were collected during a study by Moreno et al. (1994) of the same orchard but with individual trees receiving specific treatments of

no spray (ctrl), spray without insecticide (chk), bait spray with 1% cyromazine (cyr1), bait with 5% cyromazine (cyr5), and bait with 10% malathion (mal).

Table 3 gives the raw data including fruit type, types of samples, number of fruit sampled, number of fruit infested, proportion of fruit infested, total Mexican fruit fly pupae from sample, average number of pupae per infested fruit, and standard deviation of number of pupae per infested fruit. From these data, the risk (probability of a mating pair), required treatment efficacy (treatment level to have max. pest limit  $\leq 2$ ), and maximum infestation rate (proportion of fruit that may be infested to have maximum pest limit  $\leq 2$ ) have been calculated.

#### **Calculating maximum pest limit requirements based on Baker et al. (1990)**

Appendix A presents a program written in SAS language which can be used to calculate variables included in Equation (8). Appendix B presents outputs from *Mathematica*. The *Mathematica* program calculates 4 outputs.

1. The first output is the *probability of getting a mating pair* (a male and a female) from a lot having a certain maximum pest limit (*mpl*). Baker et al. (1990) use the value (*m*/lot size) for *mpl* but the calculation does not require this ratio.
2. The second output takes the maximum pests per fruit ( $\mu=\mu$ ), the treatment survival rate ( $\phi=\phi$ ), and the lot size (*N*) and calculates the *number of pests that would arrive in that lot* if every fruit were infested.
3. The third output is the *maximum permissible fruit infestation rate* that would allow not more than the maximum pest limit (*mpl*).
4. The fourth output is the *number of fruit that must be inspected* with zero pests (alive or dead), assuming 100% accuracy in inspection, to guarantee with 95% confidence that the fruit

infestation rate is below the maximum given in output 2.

Appendix C is a software package called Commodity Quarantine Treatment Statistics (CQT\_STATS). This provides user-friendly computation of Equation (8).

#### **Summary**

The calculations herein demonstrate methods that relate percentage infestation, mean larval density per fruit, and treatment efficacy for various allowable numbers of pests per load. The Vail et al. (1993) method pools the percentage infestation, mean larvae per fruit, and number of fruit per shipment as total number of pests per shipment so that the probability of a mating pair entering can then be estimated.

The Baker et al. (1990) method allows for separation of fruit infestation parameters but, like the Vail et al. (1993) method, assumes a Poisson distribution of pests among fruit (after treatment). This is probably justified if the treatment is effective to the extent that the maximum number of survivors expected is low, so that the number of survivors in a single fruit would not exceed the maximum pest limit.

The Baker et al. (1990) method is designed to estimate the maximum permissible infestation rate given the other parameters (maximum pest limit, larvae per fruit, treatment efficacy, load size), but the equations can be solved for any of these values if the others are given. For example, for load size = 250,000, larvae/fruit = 70, and infestation probability = 0.003, the required treatment efficacy to achieve  $mpl \leq 3$  is calculated as 0.0000208432 (which is about 66% more effective treatment than probit 9 level efficacy). Neither calculation method claims to give accurate estimates when the surviving pests are highly clumped such as would occur when a treatment completely fails and, even if only one fruit is infested, the survivors could found a population.



Table 2. Computed values for maximum infestation rate (proportion of fruit infested (p)) and number of fruit that should be inspected, cut, and reveal no infestation (n) from examples in Baker et al. (1990) and those calculated using *Mathematica* and formulae from Couey and Chew (1986). See text for explanation.

Load size	Larvae per fruit	Baker et al. 1990			<i>Mathematica</i>		Couey & Chew 1986		
		p	n	$\alpha^1$	p $\alpha=0.95$	n	n C=0.95	n C=0.975	
208,550	40	0.0041	887	0.948	0.004099	896.8	731.7	900	
65,650	44	0.012	307	0.948	0.1184	310.5	250.0	311.6	
53,100	12	0.0542	67	0.948	0.05366	68.5	55.3	68.1	
21,600	162	0.0099	372	0.950	0.00977	376.2	303.0	372.7	

<sup>1</sup> Calculated using "p" and "n" given in Baker et al. (1990)

Table 3. Illustration of probability of mating pair and maximum pest limit calculations using field data for oranges and mangoes from fruit producing region of Mexico. See text for explanation.

Fruit sample type location and yr	No. fruit sampled	Infested fruit		Mexican fruit fly pupae			Probability of a mating pair	Treatment efficacy (X 10 <sup>-6</sup> ) to have MPL ≤ 2	Maximum allowable infestation rate to have MPL ≤ 2
		No.	Proportion	Total	Mean	Std.dev.			
Orange on ground									
CandSVJ92	2,794	572	0.205	4,248	7.48	6.11	0.94	2.89	0.018
CandSVJ93	731	42	0.057	271	6.45	4.84	0.31	11.97	0.021
SJV 92	3,145	767	0.244	6,573	8.57	7.12	0.98	2.11	0.016
SJV 93	1,471	67	0.046	493	7.36	6.03	0.29	13.00	0.019
Mango on ground									
CandSVJ92	3,511	77	0.022	312	4.05	4.48	0.004	157.90	0.109
CandSVJ93	4,314	0	0	0	0		**	**	
SJV 92	4,320	374	0.087	1,764	4.72	4.52	0.058	34.26	0.093
SJV 93	5,031	260	0.052	1,067	4.10	4.61	0.024	58.69	0.107
Orange on tree (no treatment season)									
NL 90-1	1,000	8	0.008	51	6.38	3.80	0.01	86.24	0.022
NL 91-2	1,000	22	0.022	98	4.45	3.40	0.04	44.96	0.031
NL 91-3	1,000	65	0.065	241	3.70	2.09	0.17	18.30	0.037
NL 91-4	1,000	60	0.060	285	4.75	4.24	0.22	15.45	0.028
Orange on tree (treatment season)									
NL Ctrl	440	16	0.036	73	4.56	3.92	0.1	26.81	0.030
NL Chk	440	17	0.039	117	6.88	7.10	0.21	16.41	0.020
NL Cyr1	440	8	0.018	32	4.00	2.83	0.02	61.41	0.034
NL Cyr5	440	0	0	0	0		**	**	
NL-Mal	440	3	0.006	16	5.33	7.50	0.004	137.65	0.025



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## APPENDICES

### Appendix A - Computer Programs: Biological Statistics for Potential Approaches in Developing Commodity Quarantine Treatment, written in SAS language.

#### CQT Equation 1 - Landolt et al. (1984)

#### Probability of One or More Mating Pairs Surviving a Commercial Shipment

$$P = \sum_{x=2}^{x=\infty} (e^{-NR} (NR)^x / x!) * (1 - 0.5^{x-1}) \quad (1)$$

SAS program using data from (Landolt et al. 1984).

```
2  data;
3  put 'Landolt et al. 1984';
4  put 'Calculate probability of at least';
5  put '    one mating pair in a shipment';
6  n = 36000;      /* number of fruit in lot */;
7  r = .0001;      /* infestation rate      */;
8  nr = n*r;
9  sum=0;
10 x=2;
11 do while (x le 50);
12     xfact=1;
13     z=1;
14     do while (z le x);
15         xfact = xfact * z;
16         z = z+1;
17     end;
18     newterm=(exp(-nr)*((nr)**x)/xfact)*(1-(.5**(x-1)));
19     sum=sum+newterm;
20     x=x+1;
21 end;
22 p = sum;
23 put '                Number of fruit (N) = 'N;
24 put '                Infestation rate (R) = 'R;
25 put '                Calculated NR = 'nr;
26 put 'Probability of one or more mating pairs (P) = 'p;
27 put '-----';
28 run;
```

Landolt et al. 1984

Calculate probability of at least  
one mating pair in a shipment

Number of fruit (N) = 36000  
Infestation rate (R) = 0.0001  
Calculated NR = 3.6

Probability of one or more mating pairs (P) = 0.696725946

-----

**The following outputs were generated by the above program after changing the value of R.**

```
      Number of fruit (N) = 36000
      Infestation rate (R) = 0.00001
      Calculated NR = 0.36
Probability of one or more mating pairs (P) = 0.0271359032
-----
```

```
      Number of fruit (N) = 36000
      Infestation rate (R) = 1E-6
      Calculated NR = 0.036
Probability of one or more mating pairs (P) = 0.0003182288
-----
```

```
      Number of fruit (N) = 36000
      Infestation rate (R) = 1E-7
      Calculated NR = 0.0036
Probability of one or more mating pairs (P) = 3.2341741E-6
```



### CQT Equation 3 & 4 - Vail et al. (1993)

#### Required Level of Probit Mortality Based on the Survival Rate of the Pest Species

$$NR = -2 * (\log_e(1 - \sqrt{P})) \quad (3)$$

$$m = 1 - (NR / (i * n * s)) \quad (4)$$

SAS program using data from (Vail et al. 1993).

```
2  data;
3  /* Vail et al. 1993  Equations 3 and 4
4  I=infestation rate (#larvae/kg)
5  s=survival rate of insects infesting the host
6  n=number (or kg) of fruit
7
8  x=number of larvae in shipment (Poisson assumed)
9  P=probability of one or more mating pairs
10 NR=total number of codling moth allowed as a function of P
11 */;
12 put "-----";
13 put "lot size=20,000kg, survival rate=0.80";
14 put "98.5 Upper CL infestation rate";
15 put "P=0.05";
16 put "-----";
17 put " CHERRY";
18 I=.000004;
19 n=20000;
20 s=0.80;
21 p=0.05;
22 nr= -2 * (log (1-sqrt(p))); /* Equation #3 */;
23 put 'infestation rate ='I ;
24 put "    alpha = .95";
25 put "    NR    = " nr;
26 m=1-(NR/(I*n*s)); /* Equation #4 */;
27 put "    M    = " m;
28 if m<0 then put "    No treatment required ";
29 else do;
30 permort=m*100;
31 put "    Treatment must provide " permort " % mortality";
32 end;
33 put "-----";
34 put " NECTRINES";
35 I=.0002;
36 n=20000;
37 s=0.80;
38 p=0.05;
39 nr= -2 * (log (1-sqrt(p)));
40 put 'infestation rate ='I ;
41 put "    alpha = .95";
42 put "    NR    = " nr;
```

```

43 m=1-(NR/(I*n*s));
44 put "      M      = " m;
45 if m<0 then put "      No treatment required ";
46 else do;
47     permort=m*100;
48     put "      Treatment must provide " permort " % mortality";
49 end;
50 put "-----";
51 put " WALNUTS";
52 I=.285;
53 n=20000;
54 s=0.80;
55 p=0.05;
56 nr= -2 * (log (1-sqrt(p)));
57 put 'infestation rate ='I ;
58 put "      alpha = .95";
59 put "      NR      = " nr;
60 m=1-(NR/(I*n*s));
61 put "      M      = " m;
62 if m<0 then put "      No treatment required ";
63 else do;
64     permort=m*100;
65     put "      Treatment must provide " permort " % mortality";
66 end;
67 put "-----";
68 put "      P = 0.01";
69 put "-----";
70 put " CHERRY";
71 I=.000004;
72 n=20000;
73 s=0.80;
74 p=0.01;
75 nr= -2 * (log (1-sqrt(p)));
76 put 'infestation rate ='I ;
77 put "      alpha = .99";
78 put "      NR      = " nr;
79 m=1-(NR/(I*n*s));
80 put "      M      = " m;
81 if m<0 then put "      No treatment required ";
82 else do;
83     permort=m*100;
84     put "      Treatment must provide " permort " % mortality";
85 end;
86 put "-----";
87 put " NECTRINES";
88 I=.0002;
89 n=20000;
90 s=0.80;
91 p=0.01;
92 nr= -2 * (log (1-sqrt(p)));
93 put 'infestation rate ='I ;

```



```

94      put "      alpha = .99";
95      put "      NR      = " nr;
96      m=1-(NR/(I*n*s));
97      put "      M      = " m;
98      if m<0 then put "      No treatment required ";
99      else do;
100         permort=m*100;
101         put "      Treatment must provide " permort " % mortality";
102     end;
103     put "-----";
104     put " WALNUTS";
105     I=.285;
106     n=20000;
107     s=0.80;
108     p=0.01;
109     nr= -2 * (log (1-sqrt(p)));
110     put 'infestation rate ='I ;
111     put "      alpha = .99";
112     put "      NR      = " nr;
113     m=1-(NR/(I*n*s));
114     put "      M      = " m;
115     if m<0 then put "      No treatment required ";
116     else do;
117         permort=m*100;
118         put "      Treatment must provide " permort " % mortality";
119     end;
120 run;

```

```

-----
lot size=20,000kg, survival rate=0.80
98.5 Upper CL infestation rate
P = 0.05
-----

```

```

CHERRY
infestation rate =4E-6
      alpha = .95
      NR      = 0.5061923664
      M      = -6.909255725
      No treatment required
-----

```

```

NECTRINES
infestation rate =0.0002
      alpha = .95
      NR      = 0.5061923664
      M      = 0.8418148855
      Treatment must provide 84.18148855 % mortality
-----

```

```

WALNUTS
infestation rate =0.285
      alpha = .95
      NR      = 0.5061923664

```

```

M      = 0.99988889929
Treatment must provide 99.988889929 % mortality
-----
P = 0.01
-----
CHERRY
infestation rate =4E-6
  alpha = .99
  NR     = 0.2107210313
  M      = -2.292516114
  No treatment required
-----
NECTRINES
infestation rate =0.0002
  alpha = .99
  NR     = 0.2107210313
  M      = 0.9341496777
  Treatment must provide 93.414967771 % mortality
-----
WALNUTS
infestation rate =0.285
  alpha = .99
  NR     = 0.2107210313
  M      = 0.9999537892
  Treatment must provide 99.995378925 % mortality

```



## CQT Equation 8 - Baker et al. (1990)

### Maximum Pest Limit

$$\sum_{r=0}^m \frac{(\mu \phi N p)^r}{r!} \exp(-\mu \phi N p) - \sqrt{\alpha} = 0 \quad (8)$$

SAS program using data from (Baker et al. 1990).

```

1  /*****
2  /* Data for B. cucumis (JEE - Baker et. al. 1990) pages 13-17  */;
3  data; /* ----- Enter parameters ----- */;
4      m=3          /* max # live flies permissable (after treat)  */;
5      N=208550     /* Lot size (# fruits in shipment)  */;
6      alpha=.95    /* Confidence level (of solution)  */;
7      phi=.000032  /* Treatment efficacy  */;
8      mu=40        /* mean number of larvae per infested fruit  */;
9  /*****
10 /*SECTION A - Solve p given all other terms  */;
11 /* (maximum allowable percentage of infested fruit in lot)  */;
12 /*****
13     put "Data for B.cucumis (Baker et. al. 1990) pages 13-17";
14     put "m=3          max # live flies permissable after treat";
15     put "N=208550     Lot size (# fruits in shipment)";
16     put "alpha=.95    Confidence level ";
17     put "phi=.000032  Treatment efficacy";
18     put "mu=40        max number of larvae per infested fruit";
19     put " " ;
20     put "Section A: calc max allowable percent fruit infested (p)";
21     p=0.0000001;
22     incremen=.1;
23     lastp=p;
24 loop:if p>1 then goto err;
25     sum=0;
26     r=0;
27     do until (r>m);
28         rfact=1;
29         x=1;
30         do until (x>r);
31             rfact=rfact*x;
32             x=x+1;
33         end;
34         sum=sum+(((mu*phi*N*p)**r)/rfact)*exp(-mu*phi*N*p);
35     r=r+1;
36     end;
37     result=sum-(sqrt(alpha));
38     /*put " p = " p " result " result; */;
39     if abs(result-0)<.000001 then goto done;
40     if result<0 then do;
41         p=lastp;

```

```

42         incremen=incremen/10;
43     end;
44     lastp=p;
45     p=p+incremen;
46     goto loop;
47 done:put "Maximum permissable infestation level (p) = " p;
48     go to sectionb;
49 err:put "Solution to p not found between 1 and 0 ";
50 /*****/;
51 /* SECTION B          Solve for phi given all other terms          */;
52 /*****/;
53 sectionb:
54     put " ";
55     put "Section B: solve for phi";
56             /* p = value calculated in section A          */;
57             /*      OR assign any value you want to test*/;
58     m=3;
59     nu=40;
60     n=208550;
61     phi=.000001;
62     incremen=.01;
63     lastn=n;
64 topb:  if phi>1 then goto errorb;
65     sum=0;
66     r=0;
67     do until (r>m);
68         rfact=1;
69         x=1;
70         do until (x>r);
71             rfact=rfact*x;
72             x=x+1;
73         end;
74         sum=sum+(((mu*phi*N*p)**r)/rfact)*exp(-mu*phi*N*p));
75         r=r+1;
76     end;
77     result=sum-(sqrt(alpha));
78 /*      put " phi = " phi " Result of equation is " result  */;
79     if abs(result-0)<.000001 then goto doneb;
80     if result<0 then do;
81         incremen=incremen/10;
82         phi=lastphi;
83     end;
84     lastphi=phi;
85     phi=phi+incremen;
86     goto topb;
87 doneb: put " phi (required treatment efficacy) = " phi;
88     go to sectionc;
89 errorb: put "Solution to phi not found between .000001 and 1";
90 /*****/;
91 /* SECTION C Solve for N given p  Equ #3a - Couey & Chew 1986*/;
92 /*****/;

```



```

93  sectionc:
94      put " ";
95      put "Section C - solve for N given p= " p;
96      numf= (log(.05)) / (log (1-p));
97      put "(Couey and Chew) Number of fruits to sample = " numf;
98      /*****/;
99      /* SECTION D    Solve for N given p    Equ #3a    Baker          */;
100     /*****/;
101     put " ";
102     put "Section D - solve for N given p =" p;
103     numf= (log(1-sqrt(.95))) / (log (1-p));
104     put "(Baker) Number of fruits to sample = " numf;
105  run;

```

Data for B.cucumis (Baker et. al. 1990) pages 13-17

m=3	max # live flies permissable after treat
N=208550	Lot size (# fruits in shipment)
alpha=.95	Confidence level
phi=.000032	Treatment efficacy
mu=40	max number of larvae per infested fruit

Section A: calc max allowable percent fruit infested (p)  
Maximum permissable infestation level (p) = 0.0040992

Section B: solve for phi  
phi (required treatment efficacy) = 0.000032

Section C - solve for N given p= 0.0040992  
(Couey and Chew) Number of fruits to sample = 729.31011348

Section D - solve for N given p =0.0040992  
(Baker) Number of fruits to sample = 894.95476573

**The following outputs were generated by the above program after changing the values of m, N, alpha, phi, and mu.**

Data for B.melanotus (Baker et. al. 1990) pages 13-17

m=3	max # live flies permissable after treat
N=65650	Lot size (# fruits in shipment)
alpha=.95	Confidence level
phi=.000032	Treatment efficacy
mu=44	max number of larvae per infested fruit

Section A: max allowable percent fruit infested (p)  
Maximum permissable infestation level (p) = 0.0118381

Section B: solve for phi  
phi (required treatment efficacy) = 0.000032

Section C - solve for N given p= 0.0118381  
(Couey and Chew) Number of fruits to sample = 251.55769849

Section D - solve for N given  $p = 0.0118381$   
(Baker) Number of fruits to sample = 308.69277274

Data for *B. tryoni* (Baker et. al. 1990) pages 13-17  
 $m=3$  max # live flies permissable after treat  
 $N=53100$  Lot size (# fruits in shipment)  
 $\alpha=.95$  Confidence level  
 $\phi=.000032$  Treatment efficacy  
 $\mu=12$  max number of larvae per infested fruit

Section A: calc max allowable percent fruit infested ( $p$ )  
Maximum permissable infestation level ( $p$ ) = 0.0536661

Section B: solve for  $\phi$   
 $\phi$  (required treatment efficacy) = 0.000032

Section C - solve for N given  $p = 0.0536661$   
(Couey and Chew) Number of fruits to sample = 54.310051912

Section D - solve for N given  $p = 0.0536661$   
(Baker) Number of fruits to sample = 66.645229358

Data for *B. xanthodes* (Baker et. al. 1990) pages 13-17  
 $m=3$  max # live flies permissable after treat  
 $N=21600$  Lot size (# fruits in shipment)  
 $\alpha=.95$  Confidence level  
 $\phi=.000032$  Treatment efficacy  
 $\mu=162$  max number of larvae per infested fruit

Section A: calc max allowable percent fruit infested ( $p$ )  
Maximum permissable infestation level ( $p$ ) = 0.0097724

Section B: solve for  $\phi$   
 $\phi$  (required treatment efficacy) = 0.000032

Section C - solve for N given  $p = 0.0097724$   
(Couey and Chew) Number of fruits to sample = 305.04999471

Section D - solve for N given  $p = 0.0097724$   
(Baker) Number of fruits to sample = 374.33451355



### Couey and Chew (1986) Equation 3

#### Confidence Limits and Sample Size Determination

$$C = 1 - (1 - p_u)^n$$

This equation calculates confidence level ( $C$ ) that the true survival proportion  $p$  is less than  $p_u$  (upper limit of survival proportion) given that number of fruit sampled ( $n$ ) is known. Probit 9 assigns  $p_u = 32/1,000,000$ .

SAS programs using data from (Couey and Chew 1986).

```
1  data;
2  put 'Couey and Chew (1986): Equation #3';
3  pu = .000032;
4  n  = 50000;
5  C  = 1-(1-pu)**n;
6  put 'Question: ';
7  put 'Having tested n= ' n ' insects and found s = 0 survivor,';
8  put 'how much confidence do we have that the true survival';
9  put 'proportion p <= pu = 'pu ' ?';
10 put 'Answer: ';
11 put 'Calculated confidence level C = ' C ;
```

Couey and Chew (1986): Equation #3

Question:

Having tested n= 50000 insects and found s = 0 survivor,  
how much confidence do we have that the true survival  
proportion p <= pu = 0.000032 ?

Answer:

Calculated confidence level C = 0.7981086506

### Couey and Chew (1986) Equation 3a

$$n = \frac{\log(1-C)}{\log(1-p_u)}$$

Equation 3 rewritten to express n in terms of C and  $p_u$ .

```
13 data;
14 put 'Couey and Chew (1986): Equation #3a';
15 C  = .95;
16 pu = .000032;
17 n  = log(1-c)/log(1-pu);
18 put 'Question: ';
19 put 'How many insects must be tested such that if no survivor is';
20 put 'found, we will have 'C'% confidence that the true survival';
21 put 'proportion p <= pu = 'pu ' ?';
```

```

22     put 'Answer: ';
23     put 'Calculated number of fruit (n) = ' n ;

```

Couey and Chew (1986): Equation #3a

Question:

How many insects must be tested such that if no survivor is found, we will have 0.95 % confidence that the true survival proportion  $p \leq p_u = 0.000032$  ?

Answer:

Calculated number of fruit (n) = 93615.135674

### Couey and Chew (1986) Equation 3b

$$p_u = 1 - (1 - c)^{1/n}$$

Equation 3 rewritten to express  $p_u$  in terms of C and n.

```

25     data;
26     put 'Couey and Chew (1986): Equation #3b';
27     C = .95;
28     n = 50000;
29     pu = 1 - (1 - C)**(1/n);
30     put 'Question: ';
31     put 'Having tested n= ' n ' insects and found s = 0 survivor, ';
32     put 'we have 'C'% confidence that the true survival proportion p';
33     put 'is <= ?';
34     put 'Answer: ';
35     put 'Calculated treatment survival rate ' pu ;

```

Couey and Chew (1986): Equation #3b

Question:

Having tested n= 50000 insects and found s = 0 survivor, we have 0.95 % confidence that the true survival proportion p is <= ?

Answer:

Calculated treatment survival rate 0.0000599129

### Couey and Chew (1986) Equation 4

$$C = 1 - (1 - p_u)^n - n p_u (1 - p_u)^{n-1}$$

This equation calculates confidence level (C) that the true survival proportion p is less than  $p_u$  (upper limit of survival proportion) given that one survivor has been found out of (n) fruit sampled. Probit 9 assigns  $p_u = 32/1,000,000$ .



```

37  data;
38      put 'Couey and Chew (1986): Equation 4 (one survivors found)';
39      n = 35000;
40      pu = .000032;
41      C = 1 - (1-pu)**n - n*pu*(1-pu)**(n-1);
42      put 'Question: ';
43      put 'Having tested n= ' n ' insects and found s = 1 survivor,';
44      put 'how much confidence do we have that the true survival';
45      put 'proportion p <= pu = ' pu ' ?';
46      put 'Answer: ';
47      put 'Calculated confidence level C = ' C ;

```

Couey and Chew (1986): Equation 4 (one survivors found)

Question:

Having tested n= 35000 insects and found s = 1 survivor,  
how much confidence do we have that the true survival  
proportion p <= pu = 0.000032 ?

Answer:

Calculated confidence level C = 0.308287537

### Couey and Chew (1986) Equation 7

When more than one survivor is found the following equation is used to relate  $C$ ,  $s$  (number of survivors found),  $n$  and  $p_u$ . This equation is used to generate Couey and Chew (1986) Table 1 values.

$$\sum_{x=0}^{x=s} \frac{e^{-m} m^x}{x!} = 1 - C$$

where

$$m = np_u$$

Given any three of the four terms ( $C, s, n, p_u$ ) the fourth term can be determined.

```

49  data;
50      s = 26;                      /* Number of survivors found */;
51      C = .975;                    /* Confidence level */;
52      put 'Couey and Chew (1986): Equation 7';
53      put 'Generates Table 1 values (m)';
54      put 'With 's' survivor(s) found and 'C' confidence level,';
55      put ' calculate m (n*pu)';
56      m=1;
57      incr=10;
58      lastm=m;
59      done=0;
60      do while (done = 0);
61          sum=0;
62          x=0;
63          do while (x le s);

```

```

64      xfact =1;
65      z = 1;
66      do while (z le x);
67          xfact=xfact * z;
68          z=z+1;
69      end;
70      sum=sum+((exp(-m)*(m**x))/xfact);
71      x=x+1;
72  end;
73  result=sum-(1-C);
74  if abs(result) le .00001 then do;
75      done=1;
76  end;
77  if result < 0 then do;
78      m=lastm;
79      incr=incr/10;
80  end;
81  lastm=m;
82  m=m+incr;
83  end;
84  put 'Calculated m =' m;

```

Couey and Chew (1986): Equation 7

Generates Table 1 values (m)

With 26 survivor(s) found and 0.975 confidence level,  
calculate m (n\*pu)

Calculated m =38.096

```

86  data;
87  /*****/;
88  put 'Given any three of the four terms (s,n,pu,c)';
89  put ' in equation #7, the remaining term ';
90  put ' can be calculated.';
91  /*****/;
92  put ' ';
93  put 'Given s, n, and pu, calculate c ';
94  s = 2;          /* number of survivors */;
95  n = 75000;      /* lot size */ ;
96  pu = .00005     /* treatment survival rate */;
97  m=n*pu;
98  c=1;
99  incr=-1;
100 lastc=c;
101 done=0;
102 do while (done = 0);
103     sum=0;
104     x=0;
105     do while (x le s);
106         xfact =1;
107         z = 1;
108         do while (z le x);

```



```

109      xfact=xfact * z;
110      z=z+1;
111      end;
112      sum=sum+ ((exp(-m)*(m**x))/xfact);
113      x=x+1;
114      end;
115      result=sum-(1-c);
116      if abs(result) le .00001 then do;
117          done=1;
118      end;
119      if result < 0 then do;
120          c=lastc;
121          incr=incr/10;
122      end;
123      lastc=c;
124      c=c+incr;
125      end;
126      put 'Number of survivors      (s) = 's;
127      put 'Number of insects      (n) = 'n;
128      put 'Treatment survival rate (pu) = 'pu;
129      put 'Calculated confidence level (c) = 'c;

```

Given any three of the four terms (s,n,pu,c)  
in equation #7, the remaining term  
can be calculated.

Given s, n, and pu, calculate c

```

Number of survivors      (s) = 2
Number of insects      (n) = 75000
Treatment survival rate (pu) = 0.00005
Calculated confidence level (c) = 0.72293

```

```

131  data;
132      put 'Given s, n, and c, calculate pu';
133      s = 2;          /* number of survivors */;
134      n = 75000;      /* lot size */;
135      c = .95         /* confidence level */;
136      m=1;
137      incr=10;
138      lastm=m;
139      done=0;
140      do while (done = 0);
141          sum=0;
142          x=0;
143          do while (x le s);
144              xfact =1;
145              z = 1;
146              do while (z le x);
147                  xfact=xfact * z;
148                  z=z+1;
149              end;

```

```

150      sum=sum+ ((exp(-m)*(m**x))/xfact);
151      x=x+1;
152  end;
153  result=sum-(1-c);
154  if abs(result) le .00001 then do;
155      done=1;
156  end;
157  if result < 0 then do;
158      m=lastm;
159      incr=incr/10;
160  end;
161  lastm=m;
162  m=m+incr;
163  end;
164  pu=m/n;
165  put 'Number of survivors          (s) = 's;
166  put 'Number of insects           (n) = 'n;
167  put 'Confidence level            (c) = 'c;
168  put 'Calculated treatment survival rate (pu) = 'pu;

```

Given s, n, and C, calculate pu

```

Number of survivors          (s) = 2
Number of insects           (n) = 75000
Confidence level            (c) = 0.95
Calculated treatment survival rate (pu) = 0.0000839347

```

```

170  data;
171  put 'Given s, pu, and c, calculate n';
172      s = 2;          /* number of survivors */;
173      pu = .000032; /* treatment survival rate */;
174      c = .95          /* confidence level */;
175      m=1;
176      incr=10;
177      lastm=m;
178      done=0;
179  do while (done = 0);
180      sum=0;
181      x=0;
182      do while (x le s);
183          xfact =1;
184          z = 1;
185          do while (z le x);
186              xfact=xfact * z;
187              z=z+1;
188          end;
189          sum=sum+ ((exp(-m)*(m**x))/xfact);
190          x=x+1;
191      end;
192      result=sum-(1-c);
193      if abs(result) le .00001 then do;
194          done=1;
195      end;

```

```

196     if result < 0 then do;
197         m=lastm;
198         incr=incr/10;
199     end;
200     lastm=m;
201     m=m+incr;
202 end;
203 n=m/pu;
204 put 'Treatment survival rate      (pu) = 'pu;
205 put 'Number of survivors          (s) = 's;
206 put 'Confidence level             (c) = 'c;
207 put 'Calculated number of insects (n) = 'n;
208 run;

```

Given s, pu, and C, calculate n

```

Treatment survival rate      (pu) = 0.000032
Number of survivors          (s) = 2
Confidence level             (c) = 0.95
Calculated number of insects (n) = 196721.875

```



## Appendix B - Output from *Mathematica* calculations based on Equation (8)

### CQT Equation 8 - Baker et al. (1990)

$$\sum_{r=0}^m \frac{(\mu \phi N p)^r}{r!} \exp(-\mu \phi N p) - \sqrt{\alpha} = 0 \quad (8)$$

In[52]:=

**(\*Calculation of Max Infestation Rate & Number of Fruit Cut\*)**

In[53]:=

*(\*from Baker et al. 1990\*)*  
*(\*mu=maximum estimated larvae per fruit,*  
*phi = treatment survival (probit 9 = 32/1000000),*  
*n = lot size,*  
*mpl = maximum pest limit,*  
*ppair=probability of male and female from mpl\*)*

In[55]:=

mpl=5;  
mu=70;  
n=55296;  
phi=32/1000000;  
ppair=Sum[1+E^(-mpl))-2\*(E^(-mpl/2))]/N

Out[59]=

0.842568

In[60]:=

x=mu\*phi\*n//N

Out[60]=

123.863

In[61]:=

FindRoot[E^(-x\*p)\*Sum[((x\*p)^i)/i!,{i, 0, mpl}]  
)- (0.95^0.5) == 0, {p, .01}, MaxIterations -> 60]

Out[61]=

{p -> 0.0178309}

In[62]:=

*(\*Insert here to calculate fruit cut(mx->): from*  
*the estimate of "p" \*)*

In[63]:=

frcut=FindRoot[(1-(0.95^0.5))-(E^(-0.0178309\*mx)) == 0,  
{mx, Random[]}, MaxIterations -> 60]

Out[63]=

{mx -> 206.167}

## Output from *Mathematica* calculations

In[64]:=

**(\*Calculation of Max Infestation Rate & Number of Fruit Cut\*)**

In[65]:=

*(\*from Baker et al. 1990\*)*  
*(\*mu=maximum estimated larvae per fruit,*  
*phi = treatment survival (probit 9 = 32/1000000),*  
*n = lot size,*  
*mpl = maximum pest limit,*  
*ppair=probability of male and female from mpl\*)*

In[67]:=

mpl=3;  
mu=70;  
n=55296;  
phi=32/1000000;  
ppair=Sum[1+E^(-mpl))-2\*(E^(-mpl/2))]/N

Out[71]=

0.603527

In[72]:=

x=mu\*phi\*n//N

Out[72]=

123.863

In[73]:=

FindRoot[E^(-x\*p)\*Sum[((x\*p)^i)/i!,{i, 0, mpl}]  
)- (0.95^5) == 0, {p, .01}, MaxIterations ->60]

Out[73]=

{p -> 0.00883449

In[74]:=

*(\*Insert here to calculate fruit cut(mx->): from*  
*the estimate of "p" \*)*

In[76]:=

frcut=FindRoot[(1-(0.95^5))-(E^(-0.00883449\*mx)) == 0,  
{mx, Random[]}, MaxIterations ->60]

Out[76]=

{mx -> 416.112}

## **Appendix C - CQT\_STATS: Biological Statistics for Pest Risk Assessment in Developing Commodity Quarantine Treatment, ver. 1.2**

- The diskette for this software is inserted on the back cover.

Commodity Quarantine Treatment Statistics (CQT\_STATS) is an IBM-PC based specialty calculator created with Toolbook 4.0 (Asymetrix Corporation 1994). It provides user-friendly computation of the following features:

- Equation (1) (Landolt et al. 1984)  
Probability of One or More Mating Pairs Surviving a Commercial Shipment
- Equation (2) (Vail et al. 1993)  
Probability of One or More Mating Pairs
- Equations (3) (4) (Vail et al. 1993)  
Required Level of Probit Mortality based on the Survival Rate of the Pest Species
- Equation (5) (6) (7)  
Alternative Treatment Efficacy
- Equation (8) (Baker et al. 1990)  
Maximum Pest Limit
- Confidence Limits and Sample Size Determination (Couey and Chew 1986: Equations (3a)&(7) )
- Probit/Proportion Table - probit to proportion conversions that also calculate number of fruit to sample given a confidence level.

### **System requirements -**

To run CQT\_STATS you need

- Microsoft Windows 3.1 or higher
- a Windows-compatible computer with a 20Mhz 80386SX processor or higher
- a Windows compatible mouse or other pointing device
- a 1.44MB (3.5-inch) disk drive
- a hard disk with 8MB of free disk space (CQT\_STATS requires 200KB and runtime Toolbook will use about 4MB of storage space)
- at least 4MB of RAM (8MB recommended)
- VGA graphics card or better (SVGA recommended)



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